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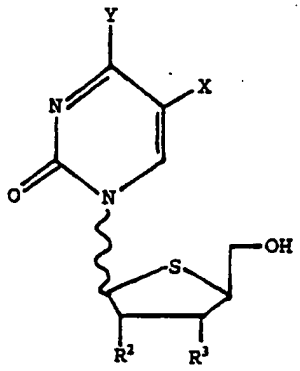

(I)

(57) Abstract

Antiviral nucleosides of formula (I), wherein: Y is hydroxy or amino; X is hydrogen, hydroxy, mercapto, halo, trifluoromethyl, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, hydroxyC_{1-3}alkyl, formyl, C_{2-6}alkenyl, C_{2-6}haloalkenyl, C_{2-6}alkynyl, C_{1-6}alkoxy, C_{1-6}alkylthio, C_{1-6}alkoxyC_{1-2}alkyl, C_{1-6}alkylthiomethyl, amino, monoC_{1-6}alkylamino, diC_{1-6}alkylamino, cyano, thiocyanate or nitro; R^2 is hydrogen and R^3 is hydroxy or hydrogen, or together R^2 and R^3 form a carbon-carbon bond; and physiologically functional derivatives thereof. Compositions containing the compounds, their use in treatment and therapy of viral disease, and processes for the production of the compounds are also provided.

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ANTIVIRAL PYRIMIDINE NUCLEOSIDES

The present invention relates to pyrimidine nucleosides and their use in medical therapy particularly for the treatment or prophylaxis of virus infections.

Of the DNA viruses, those of the herpes group are the sources of the most common viral illnesses in man. The group includes herpes simplex virus types 1 and 2 (HSV), varicella zoster virus (VZV), cytomegalovirus (CMV); Epstein-Barr virus (EBV); human herpes virus type 6 (HHV-6) and human herpes virus type 7 (HHV-7). HSV 1 and HSV 2 are some of the most common infectious agents of man. Most of these viruses are able to persist in the host's neural cells; once infected, individuals are at risk of recurrent clinical manifestations of infection which can be both physically and psychologically distressing.

HSV infection is often characterised by extensive and debilitating lesions of the skin, mouth and/or genitals. Primary infections may be subclinical although tend to be more severe than infections in individuals previously exposed to the virus. Ocular infection by HSV can lead to keratitis or cataracts thereby endangering the host's sight. Infection in the newborn, in immunocompromised patients or penetration of the infection into the central nervous system can prove fatal.

Transmission of the virus is by direct physical contact between a host and a recipient; the spread of HSV infection is therefore considered a very significant social problem, particularly as no effective vaccine is yet available.

Varicella zoster (VZV) is a herpes virus which causes chickenpox and shingles. Chickenpox is the primary disease produced in a host without immunity and in young children is usually a mild illness characterised by a vesicular rash and fever. Shingles or zoster is the recurrent form of the disease which occurs in adults who were previously infected with varicella-zoster virus. The clinical manifestions of shingles are characterised by neuralgia and a vesicular skin rash that is unilateral and dermatomal in distribution. Spread of inflammation may lead to paralysis or convulsions. Coma can occur if the meninges become affected. In immunodeficient patients VZV may disseminate causing serious or even fatal

illness. VZV is of serious concern in patients receiving immunosuppressive drugs for transplant purposes or for treatment of malignant neoplasia and is a serious complication of AIDS patients due to their impaired immune system.

In common with other herpes viruses, infection with CMV leads to a lifelong association of virus and host and, following a primary infection, virus may be shed for a number of years. Congenital infection following infection of the mother during pregnancy may give rise to clinical effects such as death or gross disease (microcephaly, hepatosplenomegaly, jaundice, mental retardation), retinitis leading to blindness or, in less severe forms, failure to thrive, and susceptibility to chest and ear infections. CMV infection in patients who are immunocompromised for example as a result of malignancy, treatment with immunosuppressive drugs following transplantation or infection with Human Immunodeficiency Virus may give rise to retinitis, pneumonitis, gastrointestinal disorders and neurological diseases. CMV infection in AIDS patients is a predominant cause of morbidity as, in 50-80% of the adult population, it is present in a latent form and can be re-activated in immunocompromised patients.

Epstein-Barr virus (EBV), a member of the herpes virus family, was discovered in 1964 and is now believed to be carried by 90% of the world's population. In the West, the main disease caused by EBV is acute or chronic infectious mononucleosis (glandular fever). Examples of other EBV or EBV associated diseases include lymphoproliferative disease which frequently occurs in persons with congenital or acquired cellular immune deficiency, X-linked lymphoproliferative disease which occurs namely in young boys, EBV-associated B-cell tumours, Hodgkin's disease, nasopharyngeal carcinoma, Burkitt lymphoma, non-Hodgkin β-cell lymphoma, immunoblastic lymphoma, thymomas and oral hairy leukoplakia. EBV infections have also been found in association with a variety of epithelial-cell-derived tumours of the upper and lower respiratory tracts including the lung.

HHV-6 has been shown to be a causative agent of infantum subitum in children and of kidney rejection and interstitial pneumonia in kidney and bone marrow transplant patients

respectively and may be associated with other diseases. There is also evidence of repression of stem cell counts in bone marrow transplant patients. HHV-7 is of undetermined disease etiology.

HBV is a viral pathogen of world-wide major importance. The virus is aetiologically associated with primary hepatocellular carcinoma and is thought to cause 80% of the world's liver cancer. In the United States more than ten thousand people are hospitalised for HBV illness each year, and an average of 250 die with fulminant disease. The United States currently contains an estimated pool of 500,000 to 1-million infectious carriers. Chronic active hepatitis generally develops in over 25% of carriers, and often progresses to cirrhosis. Clinical effects of infection with HBV range from headache, fever, malaise, nausea, vomiting, anorexia and abdominal pains. Replication of the virus is usually controlled by the immune response, with a course of recovery lasting weeks or months in humans, but infection may be more severe leading to persistent chronic liver disease outlined above.

One group of viruses which has assumed a particular importance is the retroviruses. Retroviruses form a sub-group of RNA viruses which, in order to replicate, must first 'reverse transcribe' the RNA of their genome into DNA ('transcription' conventionally describes the synthesis of RNA from DNA). Once in the form of DNA, the viral genome may be incorporated into the host cell genome, allowing it to take advantage of the host cell's transcription/ translation machinery for the purposes of replication. Once incorporated, the viral DNA is virtually indistinguishable from the host's DNA and, in this state, the virus may persist for the life of the cell.

A species of retrovirus, the lentivirus Human Immunodeficiency Virus (HIV), has been reproducibly isolated from humans with Acquired Immune Deficiency Syndrome (AIDS) or with the symptoms that frequently precede AIDS. AIDS is an immunosuppressive or immunodestructive disease that predisposes subjects to fatal opportunistic infections. Characteristically, AIDS is associated with a progressive depletion of T-cells, especially the helper-inducer subset bearing the OKT^4 surface marker. HIV is cytopathic and appears to preferentially infect

and destroy T-cells bearing the OKT^4 marker and it is now generally recognised that HIV is the aetiological agent of AIDS.

Since the discovery that HIV is the aetiological agent of AIDS, numerous proposals have been made for the anti-HIV chemotherapeutic agents that may be effective in treating AIDS. Thus, for example, European Patent Specification No. 196 185 describes 3'-azido-3'-deoxythymidine (which has the approved name zidovudine), its pharmaceutically acceptable derivatives and their use in the treatment of human retrovirus infections including AIDS and associated clinical conditions.

Examples of retroviral infections include human retroviral infections such as Human Immunodeficiency Virus (HIV), for example, HIV-1 or HIV-2, and Human T-cell Lymphotropic Virus (HTLV), for example, HTLV-I or HTLV-II, infections. AIDS and related clinical conditions such as AIDS-related complex (ARC), progressive generalized lymphadenopathy (PGL), Karposi's sarcoma, thrombocytopenic purpura, AIDS-related neurological conditions, such as multiple sclerosis or tropical paraparesis, and also anti-HIV antibody-positive and HIV-positive conditions, including such conditions in asymptomatic patients, are also conditions which may be treated by appropriate anti-viral therapy.

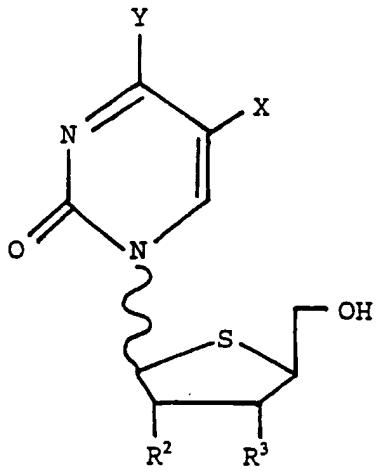
Another RNA virus which has been recognised as the causative agent of an increasingly serious international health problem is the non-A, non-B hepatitis virus. At least 80% of cases of chronic post-transfusional non-A, non-B hepatitis have been shown to be due to the virus now identified as hepatitis C and this virus probably accounts for virtually all cases of post-transfusional hepatitis in clinical settings where blood products are screened for hepatitis B. Whereas approximately half of the cases of acute hepatitis C infection resolve spontaneously over a period of months, the remainder become chronic and in many if not all such cases chronic active hepatitis ensues with the potential for cirrhosis and hepatocellular carcinoma. The structure of the hepatitis C virus genome has recently been elucidated and the virus has been characterised as a single stranded RNA virus with similarities to flaviviruses.

Coxsackie viruses belong to the enterovirus genus. They have a single stranded RNA genome contained in an icosahedral

nucleocapsid. Coxsackie virus infection is increasingly recognised as a cause of primary myocardial disease in adults and children.

Coxsackie infection is also associated with meningitis, pleurodynia, herpangia, hand-feet and mouth disease, respiratory disease, eye disease, diabetes and post-viral fatigue syndrome. In the latter case viral RNA has been detected in the muscle and in monocytes.

We have now found that certain pyrimidine L-4'-thionucleosides have activity against viruses. Such compounds include pyrimidine L-4'-thionucleosides of the following general formula (I):



wherein: Y is hydroxy or amino;

X is hydrogen, hydroxy, mercapto, halo, trifluoromethyl, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, hydroxyC_{1-3}alkyl, formyl, C_{2-6}alkenyl, C_{2-6} haloalkenyl, C_{2-6} alkynyl, C_{1-6}alkoxy, C_{1-6}alkylthio, C_{1-6}alkoxyC_{1-2}alkyl, C_{1-6}alkylthiomethyl, amino, monoC_{1-6}alkylamino, diC_{1-6}alkylamino, cyano, thiocyanate or nitro; R^2 is hydrogen and R^3 is hydroxy or hydrogen, or together R^2 and R^3 form a carbon-carbon bond;

and physiologically functional derivatives thereof.

It will be appreciated that by virtue of the definition of the group Y the compounds of formula (I) are derivatives either of uracil or of cytosine.

It will also be appreciated that the compounds of formula

(I) may exist in various tautomeric forms.

The compounds of formula (I) may exist as α- or ß-anomers; ß-anomers are preferred. It will be appreciated that the compounds of formula (I) may exist in various isomeric forms and as mixtures thereof in any proportions. The present invention includes within its scope the use of such isomeric forms or mixtures thereof, including the individual α- and β-isomers of the compound of formula (I) as well as mixtures of such isomers in any proportion.

A preferred group of compounds of the formula (I) are those in which Y is hydroxy or amino and X is hydrogen, hydroxy, mercapto, halo, trifluoromethyl, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, hydroxyC_{1-3}alkyl, formyl, C_{2-6}alkenyl, C_{2-6} haloalkenyl, C_{2-6} alkynyl, C_{1-6}alkoxy, C_{1-6}alkoxyC_{1-2}alkyl, amino, monoC_{1-6}alkylamino, diC_{1-6}alkylamino, cyano or nitro; R^2 is hydrogen and R^3 is hydroxy or hydrogen, or together R^2 and R^3 form a carbon-carbon bond; and physiologically functional derivatives thereof.

A particularly preferred group of compounds of formula (I) are those in which Y is hydroxy or amino, X is hydrogen, halo, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, C_{2-6} alkenyl, C_{2-6} haloalkenyl, C_{2-6} alkynyl, cyano or nitro; R^2 is hydrogen and R^3 is hydroxy or hydrogen, or together R^2 and R^3 form a carbon-carbon bond; and physiologically functional derivatives thereof.

In the definition of formula (I), references to alkyl groups include groups which, when they contain at least three carbon atoms may be branched or cyclic but which are preferably straight (particular alkyl groups include methyl and ethyl); references to alkenyl groups include groups which may be in the E- or Z- form or a mixture thereof and which, when they contain at least three carbon atoms, may be branched but are preferably straight; and references to alkynyl groups include groups which, when they contain at least four carbon atoms may be branched but which are preferably straight; particular alkenyl groups include vinyl and E-(1-propenyl) and particular alkynyl groups include ethynyl and prop-1-ynyl. References to halo-substituted groups include chloro, bromo, iodo and fluoro substituted groups and groups substituted with two or more halogens which may be the same or different, for example perhalo substituted groups. In

the case where the alkoxy group is part of the group alkoxyC_{1-2}alkyl, the alkoxy substituent may be attached to either the C_1- or C_2-carbon atom of the group.

Preferred compounds of the formula (I) include those in which the group X is methyl, C_{2-4} alkyl or haloalkenyl preferably C_{2-3} alkyl, C_{3-4} alkenyl or alkynyl, or halovinyl. Preferred haloalkenyl groups are straight chain haloalkenyl groups having a single halogen group on the terminal carbon. Also preferred are haloalkenyl groups having a double bond in the 1-position. Of such compounds, those having a 2-halovinyl group which is in the E configuration are preferred. Particular halovinyl groups include E-(2-bromovinyl).

Particularly preferred compounds of formula (I) are those:

(i) wherein Y is amino;

(ii) wherein R^2 and R^3 form a carbon-carbon bond;

(iii) wherein Y is amino, X is hydrogen or halo and R^2 and R^3 form a carbon-carbon bond.

Particular compounds of the invention are compounds of formula (I) and physiologically acceptable derivatives thereof wherein the pyrimidine base is selected from:

Uracil;
Cytosine;
Thymine;
5-Iodouracil;
5-Bromouracil;
5-Chlorouracil;
5-Fluorouracil;
5-Iodocytosine;
5-Bromocytosine;
5-Chlorocytosine;
5-Fluorocytosine;
5-Ethynyluracil;
5-Prop-1-ynyluracil;
5-Vinyluracil;
E-5-(2-Bromovinyl)uracil;
E-5-(1-Propenyl)uracil;
5-Ethyluracil;
5-Trifluoromethyluracil;

E-5-(2-Bromovinyl)cytosine; or
5-Propyluracil.

Compounds of formula (I) having the beta configuration which are of special interest as antiviral agents are:
2'-deoxy-4'-thio-L-uridine,
2'-deoxy-4'-thio-L-cytidine,
2'-deoxy-5-fluoro-4'-thio-L-cytidine
2'-deoxy-5-methyl-4'-thio-L-uridine,
5-(2-chloroethyl)-2'-deoxy-4'-thiouridine;
5-nitro-2'-deoxy-4'-thiouridine;
5-amino-2'-deoxy-4'-thiouridine;
5-methylamino-2'-deoxy-4'-thiouridine;
E-5-(2-bromovinyl)-2'-deoxy-4'-thio-L-uridine,
2'-deoxy-5-iodo-4'-thio-L-uridine,
5-bromo-2'-deoxy-4'-thio-L-uridine,
5-chloro-2'-deoxy-4'-thio-L-uridine,
2'-deoxy-5-ethyl-4'-thio-L-uridine,
2'-deoxy-5-prop-1-ynyl-4'-thio-L-uridine,
2'-deoxy-5-fluoro-4'-thio-L-uridine,
2'-deoxy-5-trifluoromethyl-4'-thio-L-uridine,
2'-deoxy-5-ethynyl-4'-thio-L-uridine,
2'-deoxy-5-E-(2-bromovinyl)-4'-thio-L-cytidine,
2'-deoxy-5-propyl-4'-thio-L-uridine,
E-2'-deoxy-5-(propen-1-yl)-4'-thio-L-uridine,
1-(2,3-didehydro-2,3-dideoxy-4-thio-L-ribofuranosyl)-5-methyluracil,
1-(2,3-dideoxy-4-thio-L-ribofuranosyl)-5-methyluracil,
5-bromo-2'3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine,
5-chloro-2',3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine, and
2',3'-didehydro-2',3'-dideoxy-5-iodo-4'-thio-β-L-cytidine.

Especially preferred compounds of the invention are:
2',3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine, and
2',3'-didehydro-2',3'-dideoxy-5-fluoro-4'-thio-β-L-cytidine.

These compounds have particularly potent activity against HIV and HBV.

The above-mentioned derivatives include the pharmaceutically acceptable salts; esters and salts of esters, or any other compound which, upon administration to a human subject, is capable of providing (directly or indirectly) the antivirally

active metabolite or residue thereof.

Preferred mono- and di-esters according to the invention include carboxylic acid esters in which the non-carbonyl moiety of the ester grouping is selected from straight or branched chain alkyl, (methyl, n-propyl,, n-butyl or t-butyl); cyclic alkyl (e.g. cyclohexyl); alkoxyalkyl (e.g. methoxymethyl), carboxyalkyl (e.g. carboxyethyl), aralkyl (e.g. benzyl), aryloxyalkyl (e.g. phenoxymethyl), aryl (e.g. phenyl optionally substituted by halogen, C_{1-4} alkyl or C_{1-4} alkoxy or amino); sulphonate esters such as alkyl- or aralkyl- sulphonyl (e.g. methanesulphonyl); mono-, di- or tri-phosphate esters which may or may not be blocked, amino acids esters (e.g. L-valyl or L-isoleucyl esters) and nitrate esters. With regard to the above-described esters, unless otherwise specified, any alkyl moieties present in such esters advantageously contain 1 to 18 carbon atoms, particularly 1 to 4 carbon atoms, in the case of straight chain alkyl groups, or 3 to 7 carbon atoms in the case of branched or cyclic alkyl groups. Any aryl moiety present in such esters advantageously comprises a phenyl group. Any reference to any of the above compounds also includes a reference to a physiologically acceptable salt thereof.

Salts according to the invention which may be conveniently used in therapy include physiologically acceptable base salts, eg derived from an appropriate base, such as alkali metal (e.g. sodium), alkaline earth metal (e.g. magnesium) salts, ammonium and NR_4 (wherein R is C_{1-4} alkyl) salts. When Y represents an amino group, salts include physiologically acceptable acid addition salts, including the hydrochloride and acetate salts.

Derivatives of the compounds of the formula I include the corresponding sulphones and sulphoxides.

Such nucleosides and their derivatives will be hereinafter referred to as the compounds according to the invention. The term "active ingredient" as used hereafter, unless the context requires otherwise, refers to a compound according to the invention.

The present invention further includes:

a) compounds according to the invention for use in a method of treatment or therapy of the human or animal

body.

b) compounds according to the invention for use in the treatment or prophylaxis of viral infections, eg. HBV, HIV, Hepatitis C and herpes virus infections such as those mentioned above and more particularly HSV, HHV-6, VZV, EBV or CMV infections.

c) a method for the treatment or prophylaxis of a virus infection such as those mentioned above in a mammal including man, eg. HBV, HIV, Hepatitis C and herpes virus infections such as those mentioned above and more particularly HSV, HHV-6, VZV, EBV or CMV infections, which comprises treating a subject with an effective non-toxic amount of a compound according to the invention.

d) use of a compound according to the invention in the manufacture of a medicament for use in the treatment or prophylaxis of a virus infection, such as those mentioned above, eg. HBV, HIV, Hepatitis C and herpes virus infections such as those mentioned above and more particularly HSV, HHV-6, VZV, EBV or CMV infections.

Examples of clinical conditions which may be treated in accordance with the invention include those which have been discussed in the introduction hereinbefore and in particular those caused by infections of HIV, HBV, Hepatitis C, HSV 1 and 2, HHV-6, VZV, EBV or CMV.

The compounds according to the invention are particularly useful for the treatment of humans diagnosed as HIV positive or anti-HIV antibody-positive and especially useful for the treatment of patients who are immunocompromised, for example, as a result of malignancy, treatment with immunosuppressive drugs following transplantation or humans suffering from AIDS, AIDS related complex (ARC) or progressive generalised lymphadenopathy (PGL).

The compounds according to the invention may be administered to mammals including humans by any route appropriate to the condition to be treated, suitable routes including oral, rectal, nasal, topical (including buccal and sublingual), vaginal

and parenteral (including subcutaneous, intramuscular, intravenous, intradermal, intrathecal and epidural). It will be appreciated that the preferred route may vary with, for example, the condition of the recipient.

For each of the above-indicated utilities and indications the amount required of the individual active ingredients will depend upon a number of factors including the severity of the condition to be treated and the identity of the recipient and will ultimately be at the discretion of the attendant physician. In general, however, for each of these utilities and indications, a suitable, effective dose will be in the range 0.05 to 250 mg per kilogram body weight of recipient per day, preferably in the range 0.1 to 100 mg per kilogram body weight per day and most preferably in the range 0.5 to 20 mg per kilogram body weight per day; an optimum dose is about 2 to 5 mg per kilogram body weight per day (unless otherwise indicated all weights of active ingredient are calculated as the parent compound; for salts and esters thereof the figures would be increased proportionately.) The desired dose may if desired be presented as two, three, four or more sub-doses administered at appropriate intervals throughout the day. These sub-doses may be administered in unit dosage forms, for example, containing 10 to 1000 mg, preferably 20 to 500 mg and most preferably 100 to 400 mg of active ingredient per unit dosage form.

While it is possible for the compounds to be administered alone it is preferable to present them as pharmaceutical formulations. The formulations of the present invention comprise at least one active ingredient, as above defined, together with one or more acceptable carriers thereof and optionally other therapeutic ingredients. The carrier(s) must be "acceptable" in the sense of being compatible with the other ingredients of the formulation and not deleterious to the recipients thereof.

The formulations include those suitable for oral, rectal, nasal, topical (including buccal and sublingual), vaginal or parenteral (including subcutaneous, intramuscular, intravenous, intradermal, intrathecal and epidural) administration. The formulations may conveniently be presented in unit dosage form and may be prepared by any of the methods well known in the art

of pharmacy. Such methods include the step of bringing into association the active ingredient with the carrier which constitutes one or more accessory ingredients. In general the formulations are prepared by uniformly and intimately bringing into association the active ingredient with liquid carriers or finely divided solid carriers or both, and then, if necessary, shaping the product.

Formulations of the present invention suitable for oral administration may be presented as discrete units such as capsules, cachets or tablets each containing a predetermined amount of the active ingredient; as a powder or granules; as a solution or a suspension in an aqueous liquid or a non-aqueous liquid; or as an oil-in-water liquid emulsion or a water-in-oil liquid emulsion. The active ingredient may also be presented as a bolus, electuary or paste.

A tablet may be made by compression or moulding, optionally with one or more accessory ingredients. Compressed tablets may be prepared by compressing in a suitable machine the active ingredient in a free-flowing form such as a powder or granules, optionally mixed with a binder (e.g. povidone, gelatin, hydroxypropylmethyl cellulose), lubricant, inert diluent, preservative, disintergrant (e.g. sodium starch glycolate, cross-linked povidone, cross-linked sodium carboxymethyl cellulose), surface-active or dispersing agent. Moulded tablets may be made by moulding in a suitable machine a mixture of the powdered compound moistened with an inert liquid diluent. The tablets may optionally be coated or scored an may be formulated so as to provide slow or controlled release of the active ingredient therein using, for example, hydroxpropylmethylcellulose in varying proportions to provide desired release profile.

Formulations suitable for oral use may also include buffering agents designed to neutralise stomach acidity. Such buffers may be chosen from a variety of organic or inorganic agents such as weak acids or bases admixed with their conjugated salts.

A capsule may be made by filling a loose or compressed powder on an appropriate filling machine, optionally with one or more additives. Examples of suitable additives include binders

such as povidone; gelatin, lubricants, inert diluents and disintegrants as for tablets. Capsules may also be formulated to contain pellets or discrete sub-units to provide slow or controlled release of the active ingredient. This can be achieved by extruding and spheronising a wet mixture of the drug plus an extrusion aid (for example microcrystalline cellulose) plus a diluent such as lactose. The spheroids thus produced can be coated with a semi-permeable membrane (for example ethyl cellulose, Eudragit WE30D) to produce sustained release properties.

An edible foam or whip formulation ideally comprises; 50-70% of an edible oil, particularly a vegetable oil, including corn oil, peanut oil, sunflower oil, olive oil and soybean oil; 2-10% of one or more surfactants particularly lecithin, polyols, polyol polymer esters including glyceryl fatty acid esters, polyglyceryl fatty and acid esters (e.g. decaglycerol tetraoleate), or sorbitan fatty acid esters (e.g. sorbitan monostearate); 1-4% of a propellant which is suitable for ingestion, notably a compressed gas propellant especially nitrogen, nitrous oxide or carbon dioxide, or a gaseous hydrocarbon especially propane, butane or isobutane; 0.5-30% of one or more viscosity modifiers of particle size in the range 10-50 microns in diameter, particularly powdered sugars or colloidal silicon dioxide; and optionally 0.5-1% of one or more suitable, non-toxic colourings, flavourings or sweetners. The active ingredient is preferably present in such formulations in a concentration of 10-46%, advantageously 30%. An edible foam or whip formulation as described above may be prepared in a conventional manner, for example by mixing the edible oil, surfactant(s) and any other soluble ingredients, adding the viscosity modifier(s) and milling the mixture to form a uniform dispersion and suspension. The active ingredient is blended into the milled mixture until evenly dispersed. Finally, a metered quantity of propellant is incorporated to the mixture after said mixture has been measured into a suitable dispensing container.

Pharmaceutical formulations for topical administration according to the present invention may be formulated as an ointment, cream, suspension, lotion, powder, solution, paste,

gel, spray, aerosol or oil. Alternatively, a formulation may comprise a dressing such as a bandage or adhesive plaster impregnated with active ingredients and optionally one or more excipients or diluents.

Compositions suitable for transdermal administration may be presented as discrete patches adapted to remain in intimate contact with the epidermis of the recipient for a prolonged period of time. Such patches suitably contain the active compound 1) in an optionally buffered, aqueous solution or 2) dissolved in an adhesive or 3) dispersed in a polymer. A suitable concentration of the active compound is about 1% to 35%, prefrably about 3% to 15%. As one particular possibility, the active compound may be delivered from the patch by iontophoresis as generally described in Pharmaceutical Research, 3(6), 318 (1986).

For infections of the eye or other external tissues, e.g., mouth and skin, the formulations are preferably applied as a topical ointment or cream containing the active ingredient in an amount of, for example, 0.075 to 20% w/w, preferably 0.2 to 15% w/w and most preferably 0.5 to 10% w/w. When formulated in an ointment, the active ingredients may be employed with either a paraffinic or a water-miscible ointment base. Alternatively, the active ingredients may be formulated in a cream with an oil-in-water cream base.

If desired, the aqueous phase of the cream base may include, for example, at least 30% w/w of a polyhydric alcohol, i.e. an alcohol having two or more hydroxyl groups such as propylene glycol, butane-1,3-diol, mannitol, sorbitol, glycerol and polyethylene glycol and mixtures thereof. The topical formulations may desirably include a compound which enhances absorption or penetration of the active ingredient through the skin or other affected areas. Examples of such dermal penetration enhancers include dimethylsulphoxide and related analogues.

The oily phase of the emulsions of this invention may be constituted from known ingredients in a known manner. While this phase may comprise merely an emulsifier (otherwise known as an emulgent), it desirably comprises a mixture of at least one

emulsifier with a fat or an oil or with both a fat and an oil. Preferably, a hydrophilic emulsifier is included together with a lipophilic emulsifier which acts as a stabilizer. It is also preferred to include both an oil and a fat. Together, the emulsifier(s) with or without stabilizer(s) make up the so-called emulsifying wax, and the wax together with the oil and/or fat make up the so-called emulsifying ointment base which forms the oily dispersed phase of the cream formulations.

Emulgents and emulsion stabilizers suitable for use in the formulation of the present invention include Tween 60, Span 80, cetostearyl alcohol, myristyl alcohol, glyceryl mono-stearate and sodium lauryl sulphate.

The choice of suitable oils or fats for the formulation is based on achieving the desired cosmetic properties, since the solubility of the active compound in most oils likely to be used in pharmaceutical emulsion formulations is very low. Thus the cream should preferably be a non-greasy, non-staining and washable product with suitable consistency to avoid leakage from tubes or other containers. Straight or branched chain, mono- or dibasic alkyl esters such as di-isoadipate, isocetyl stearate, propylene glycol diester of coconut fatty acids, isopropyl myristate, decyl oleate, isopropyl palmitate, butyl stearate, 2-ethylhexyl palmitats or a blend of branched chain esters known as Crodamol CAP may be used, the last three being preferred esters. These may be used alone or in combination depending on the properties required. Alternatively, high melting point lipids such as white soft paraffin and/or liquid paraffin or other mineral oil can be used.

Formulations suitable for topical administration to the eye also include eye drops wherein the active ingredient is dissolved or suspended in a suitable carrier, especially an aqueous solvent for the active ingredient. The active ingredient is preferably present in such formulations in a concentration of 0.5 to 20%, advantageously 0.5 to 10% particularly about 1.5% w/w.

Formulations suitable for topical administration in the mouth include lozenges comprising the active ingredient in a flavoured basis, usually sucrose and acacia or tragacanth; pastilles comprising the active ingredient in an inert basis such

as gelatin and glycerin, or sucrose and acacia; and mouth-washes comprising the active ingredient in a suitable liquid carrier.

Formulations for rectal administration may be presented as a suppository with a suitable base comprising for example cocoa butter or a salicylate.

Formulations suitable for nasal administration wherein the carrier is a solid include a coarse powder having a particle size for example in the range 20 to 500 microns which is administered in the manner in which snuff is taken, i.e. by rapid inhalation through the nasal passage from a container of the powder held close up to the nose. Suitable formulations wherein the carrier is a liquid, for administration as for example a nasal spray or as nasal drops, include aqueous or oily solutions of the active ingredient.

Formulations suitable for vaginal administration may be presented as pessaries, tampons, creams, gels, pastes, foams or spray formulations containing in addition to the active ingredient such carriers as are known in the art to be appropriate.

Formulations suitable for parenteral administration include aqueous and non-aqueous sterile injection solutions which may contain anti-oxidants, buffers, bacteriostats and solutes which render the formulation isotonic with the blood of the intended recipient; and aqueous and non-aqueous sterile suspensions which may include suspending agents and thickening agents, and liposomes or other microparticulate systems which are designed to target the compound to blood components or one or more organs. The formulations may be presented in unit-dose or multi-dose containers, for example sealed ampoules and vials, and may be stored in a freeze-dried (lyophilized) condition requiring only the addition of the sterile liquid carrier, for example water for injections, immediately prior to use. Injection solutions and suspensions may be prepared extemporaneously from sterile powders, granules and tablets of the kind previously described.

Preferred unit dosage formulations are those containing a daily dose or unit, daily sub-dose, as herein above recited, or an appropriate fraction thereof, of an active ingredient.

It should be understood that in addition to the ingredients

particularly mentioned above the formulations of this invention may include other agents conventional in the art having regard to the type of formulation in question, for example those suitable for oral administration may include flavoring agents.

The compounds according to the invention may be employed alone or in combination with other therapeutic agents for the treatment of the above infections or conditions. Combination therapies according to the present invention comprise the administration of at least one compound of the formula (I) or a physiologically functional derivative thereof and at least one other pharmaceutically active ingredient. The active ingredient(s) and pharmacologically active agents may be administered together or separately and, when administered separately this may occur simultaneously or sequentially in any order. The amounts of the active ingredient(s) and pharmacologically active agent(s) and the relative timings of administration will be selected in order to achieve the desired combined therapeutic effect. Preferably the combination therapy involves the administration of one compound of the formula (I) or a physiologically functional derivative thereof and one of the agents mentioned herein below.

Examples of such further therapeutic agents include agents that are effective for the treatment of HIV infections or associated conditions such as 3'-azido-3'-deoxythymidine (zidovudine), other 2',3'-dideoxynucleosides such as 2',3'-dideoxycytidine, 2',3'-dideoxyadenosine and 2',3'-dideoxyinosine, carbovir, pentoxifylline, N-acetylcysteine, procysteine, a-trichosanthin, acyclic nucleosides (for example, acyclovir), 2',3'-didehydrothymidine, protease inhibitors such as N-tert-butyl-dechydro-2-[2(R)-hydroxy-4-phenyl-3-(S)-[[N-(2-quinolycarbonyl)-L-asparginyl]-butyl]-(4aS,8aS)-isoquinoline-3-(S)-carboxamide (RO 31-8959), oxathiolan nucleoside analogues such as cis-1-(2-hydroxymethyl)-1,3-oxathiolan-5-yl)cytosine (BCH-189) or cis-1-(2-(hydroxymethyl)-1,3-oxathiolan-5-yl)-5-fluoro-cytosine, 3'-deoxy-3'-fluorothymidine, 2',3'-dideoxy-5-ethynyl-3'-fluorouridine, 5-chloro-2'3-dideoxy-3'-fluorouridine, Ribavirin, 9-[4-hydroxy-2-(hydroxymethyl)but-1-yl]guanine(H2G), TAT inhibitors such as 7-chloro-5-(2-pyrryl)-3H-1,4-

benzodiazapin-2(H)-one (RO5-3335) or 7-chloro-1,3-dihydro-5-(1H-pyrrol-2-yl)-3H-1,4-benzodiazapin-2-amine (RO24-7429), interferons such as α-interferon, renal excretion inhibitors such as probenecid, nucleoside transport inhibitors such as dipyridamole, phosphonoformic acid, as well as immunodulators such as interleukin II, granulocyte macrophage colony stimulating factors, erythropoetin, soluble CD_4 and genetically engineered derivatives thereof. Examples of such further therapeutic agents which are effective for the treatment of HBV infections include carbovir, oxathiolan nucleoside analogues such as cis-1-(2-hydroxymethyl)-1,3-oxathiolan-5-yl)-cytosine (BCH-189) or cis-1-(2-(hydroxymethyl)-1,3-oxathiolan-5-yl-5-fluoro-cytosine, 2',3'-dideoxy-5-ethynyl-3'-fluorouridine, 5-chloro-2',3'-dideoxy-3'-fluorouridine, 1-(β-D-arabinofuranosyl)-5-propynyluracil, acyclovir and interferons, such as α-interferon. Examples of further therapeutic agents which are effective for the treatment of herpes virus infections are acyclovir, 9-[4-hydroxy-2-(hydroxymethyl)butyl]guanine (H2G), 9-[4-hydroxy-3-(hydroxymethyl)-but-1-yl)guanine (penciclovir), famciclovir, the 6-deoxy-diacetate ester of penciclovir, BVaraU, 1-(β-D-arabinofuranosyl-5-propynyluracil, 2-[(2-amino-1,6-dihydro-6-oxo-9H-purin-9-yl)methoxy]ethyl L-valinate, phosphonoformic acid and phosphonoacetic acid, Ganciclovir, (S)-1-(3-hydroxy-2-phosphonylmethoxypropyl)-cytosine (HPMPC), Oxetanocin G, 2'-deoxy-5-iodouridine, E-5-2-bromovinyl-2'-deoxy-uridine (BVDU) and 9-(3-hydroxypropoxy)guanine. Further compounds include those disclosed in EP-A-0 409 575 and EP-A-0 427 777.

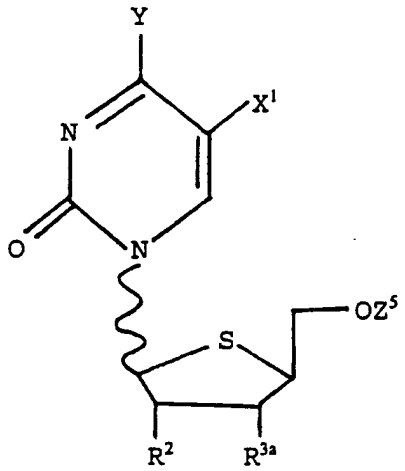
More preferably the combination therapy involves the administration of one of the above-mentioned agents and a compound within one of the preferred or more-preferred sub-groups within formula (I) as described above. Most preferably the combination therapy involves the joint use of one of the above named agents together with one of the compounds of formula (I) specifically named herein.

The compounds of formula (I) may be produced by various methods known in the art of organic chemistry in general and nucleoside synthesis in particular. Starting materials are either known and readily available from commercial sources or may

themselves be produced by known and conventional techniques.

The present invention further provides a process for producing a compound of formula (I) as hereinbefore defined which process comprises:

A) reacting a compound of formula (II)



(II)

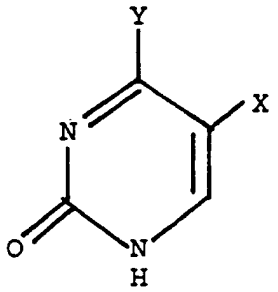
wherein X^1 is a precursor for the group X as defined in relation to formula (I);

Y and R^2 are as defined in relation to formula (I);

R^{3a} either forms a carbon-carbon bond with R^2 or when R^2 is H, R^{3a} is hydrogen, hydroxy or a group OZ^3 where Z^3 is a hydroxyl protecting group; and

Z^5 is hydrogen or a hydroxyl-protecting group, with a reagent or reagents serving to convert the group X^1 to the desired group X;

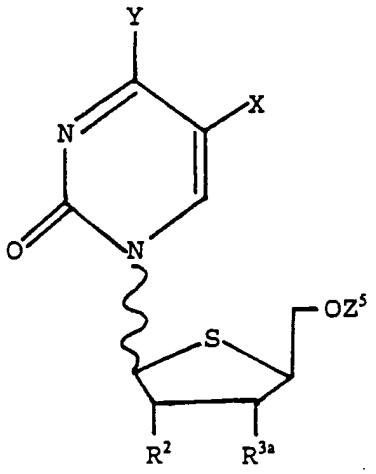
B) reacting a compound of formula (III)



(III)

wherein X and Y are as defined in relation to formula (I) or a protected form thereof with a 4-thio sugar compound serving to introduce the 4-thio sugar moiety, or a protected form thereof, at the 1-position of the compound of formula (III); or

C) reacting a compound of formula (IV)



(IV)

wherein X and Y are as defined in relation to formula (I), Z^5 is a hydroxyl protecting group or hydrogen; R^2 and R^{3a} are as defined above wherein at least one of R^{3a} and Z^5 represents a precursor group for the group(s) R^3 and/or R^5 in formula (I) under conditions or with a reagent serving to convert the groups R^{3a} and/or Z^5 into the respective groups R^3 and/or H.

Where necessary or desired, one or more of the following further steps may be additionally performed in any desired or necessary order:

a) removing each of the protecting groups,

b) converting a compound of formula (I) or a protected form thereof into a further compound of formula (I) or a protected form thereof,

c) converting the compound of formula (I) or a protected form thereof into a physiologically acceptable derivative of the compound of formula (I) or a protected form thereof,

d) converting a physiologically acceptable derivative of the compound of formula (I) or a protected form thereof into the compound of formula (I) or a protected form, thereof,

e) converting a physiologically acceptable derivative of the compound of formula (I) or a protected form thereof into another physiologically acceptable derivative of the compound of formula (I) or a protected form thereof,

f) performing an anomerisation reaction in order to convert an

α-anomer of a compound of formula (I) into a β-amoner or to convert a β-anomer of a compound of formula (I) into an α-anomer, and

g) where necessary, separating the α and ß anomers of the compound of formula I or a protected derivative thereof or of a physiologically acceptable derivative of a compound of formula (I) or a derivative thereof.

The term "4-thio sugar compound" is used herein to denote a compound containing the 4-thio-L-ribofuranose ring wherein the 5-hydroxyl group thereof is optionally protected, the 1-position is optionally substituted by a leaving group and the 2- and 3-positions thereof are either the groups R^2 and R^3 or a derivative thereof, the derivatives of R^3 including protected hydroxyl groups.

With regard to the protecting groups referred to herein, including the groups Z^3 and Z^5, it will be appreciated that the particular nature of such groups will be dependent on the identity and nature of the particular group(s) to be protected and will therefore be selected in accordance with conventional techniques. Examples of protecting groups that may be generally used include acyl groups. Acyl groups include for example C_{1-6} alkanoyl groups, e.g. acetyl, or aroyl groups, e.g. benzoyl optionally substituted with one or more C_{1-4}alkyl (particularly methyl), halo, nitro or amino groups. Preferred substituted aroyl groups include toluoyl and p-nitrobenzoyl groups. Other protecting groups include ether groups such as tri-C_{1-6}alkylsilyl (e.g. trimethylsilyl) or tert-butyl diphenylsilyl; or arylmethyl groups such as a triphenylmethyl group or a benzyl group optionally substituted on the phenyl ring by one or more halogen atoms, C_{1-4} alkyl eg. methyl, C_{1-4} haloalkyl, C_{1-4} alkoxy, nitro or amino groups.

Protection of hydroxy groups with trialkylsilyl, eg. trimethylsilyl, groups on the pyrimidine ring is conveniently achieved by reaction with (a) chlorotrimethylsilane together with triethylamine or with (b) hexamethyldisilazane, optionally together with chlorotrimethylsilane and/or ammonium sulphate.

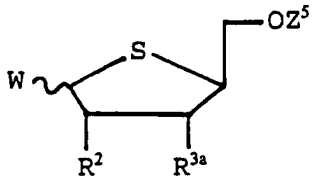
The above groups may be removed in conventional manner, for example the acyl groups being removed advantageously under basic

conditions (e.g. using sodium methoxide), the silyl ether groups being removed advantageously under aqueous or acidic conditions (e.g. using aqueous methanol to remove trimethylsilyl groups) and the arylmethyl groups being removed advantageously under reducing conditions.

Process A may be effected by, for example, the means described below.

Process B may be effected, for example, by

a) reaction of the compound of formula (III), or a protected form thereof, with a 4-thio sugar compound of formula (V):

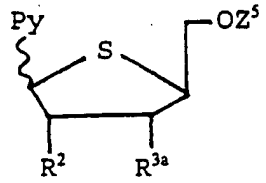


(V)

wherein R^2, R^{3a} and Z^5 are as defined above and W is a leaving group, for example halogen, e.g. chloro, acyloxy (e.g. C_{1-6} alkanoyloxy such as acetoxy), or an optionally substituted S-benzyl group of the formula -S-CH_2-Ar where Ar is an optionally substituted aryl group, for example optionally substituted phenyl or toluyl. Optional substituents of the aryl groups include one or more halogen atoms, C_{1-4} alkyl eg. methyl, C_{1-4} haloalkyl, C_{1-4} alkoxy, nitro or amino groups. Ar is preferably a 4-methoxyphenyl group. In formula (V), the groups R^{3a} and Z^5 are preferably hydroxyl protecting groups, particularly benzyl, toluoyl or p-nitrotoluoyl groups. The reaction may be performed using standard methods including the use of a Lewis Acid catalyst such as mercuric chloride or bromide or stannic chloride or trimethylsilyltrifluoromethane-sulphonate in solvents such as acetonitrile, 1-2 dichloroethane, dichloromethane, chloroform or toluene at reduced, ambient or elevated temperature such as from -78°C to reflux; or

b) reaction of the compound of formula (III), or a

protected form thereof, with a compound of formula (VI)



(VI)

wherein R^2, R^{3a} and Z^5 are as defined above and Py represents a pyrimidine base in the presence of a silylating agent such as N,O-bis-(trimethylsilyl)acetamide and in the presence of a Lewis Acid catalyst such as trimethylsilyltrifluoromethane sulphotonate in a solvent such as acetonitrile. In the compound of formula (VI), Py is preferably uracil or thymine.

The 4-thio-sugar compound of formula (V) may be produced by conventional methods prior to coupling with the base or derived by modification of another sugar moiety which is already part of a nucleoside. Particular methods are as described in the Examples.

The compound of formula (VI) may also be produced by coupling a compound of formula (V) with an appropriate base. Particular methods are as described in the Examples. For the preparation of compounds of the formula (I) the base (III) is preferably modified to protect the 2-carbonyl and 4-amine groups by silylation. Suitable silylating agents include bis-(trimethylsilyl) acetamide. Silylation is conducted in a suitable solvent, for example acetonitrile. The reaction may be conducted at from about 20°C to 100°C and is preferably preformed at an elevated temperature, e.g. about 50° to 100°C, e.g. about 80°C.

The compound of formula (V) is reacted with the protected base in the presence of a Lewis acid catalyst such as those mentioned above and, as a co-catalyst a N-halosuccinimide, eg. N-iodosuccinimide or N-bromosuccinimide. The Lewis acid is preferably trimethylsilyltrifluoromethanesulphonate. The solvent may be any suitable solvent including chlorinated solvents such as chloroform, dichloromethane, 1-2-dichloromethane but is preferably acetonitrile.

The Lewis acid and N-halosuccinimide are preferably used in

equimolar proportions, although a range of from 1:5 to 5:1 molar ratio may be used. Desirably, the ratio of Lewis acid to compound of formula (V) is 1:1.

When the nucleoside of formula (VI) is protected, it may be deprotected using standard de-esterification reactions, eg by reaction with a base (organic or inorganic) in a suitable solvent such as alcohol (eg. methanol, ethanol or propanol). The final product will be an anomeric mixture which may be separated by standard techniques such as fractional crystallisation or chromatography.

Particular methods for producing the compounds of formula (I) in accordance with the above processes will be described below and these may be combined in order to produce further compounds within formula (I), in accordance with Process A above.

Reference may be made to the following texts:

Synthetic Procedures in Nucleic Acid Chemistry, Eds. W.W. Zorbach R.S. Tipson, Vol. 1, Interscience, 1973;

Nucleic Acid Chemistry - Improved and New Synthetic Procedures, Methods and Techniques, Eds. L.B. Townsend and R.S.Tipson, Parts 1 and 2, Wiley-Interscience, 1978 and Part 3, Wiley-Interscience, 1986;

Nucleoside Analogues-Chemistry, Biology and Medical Applications Eds R.T Walker, E. De Clercq & F. Eckstein, NATO Advanced Study Institutes Series, Plenum Press, 1979; Basic Principles in Nucleic Acid Chemistry, Eds. P.O.P Ts'O, Academic Press, 1974.

The following techniques are particularly convenient:

<u>X is halogen</u>

5-Halopyrimidines are commercially available and may be coupled to the 4-thiosugar compound by conventional techniques, for instance by reacting a protected 5-halopyrimidine with a protected 4-thio sugar compound having a leaving group in the 1-position. The leaving group on the 4-thio sugar compound may be a halogen, benzylthio or preferably acetate group.

Reaction of the protected 4-thio sugar compound with the protected 5-halopyrimidine is conducted under conventional conditions using Lewis Acid catalysis such as by treatment with mercuric chloride or mercuric dibromide with cadmium carbonate or

with stannic chloride, or preferably trimethylsilyl-trifluoromethane sulphonate in toluene, acetonitrile, dichloromethane or 1,2-dichloroethane, as solvent followed by treatment as necessary with aqueous methanol (which also serves to remove the protecting groups from any hydroxyls on the pyrimidine ring).

Protecting groups may be removed by conventional techniques, for instance trimethylsilyl groups may be removed from hydroxyl groups on the pyrimidine ring by treatment with aqueous methanol, benzyl groups are removed from the hydroxyl groups on the 4-thio sugar compound by treatment with boron trichloride in dichloromethane at -78°C, and p-toluyl groups are removed from the hydroxyl groups on the sugar by treatment with sodium methoxide in methanol at room temperature.

Alternatively the 5-halo substituent may be introduced into the pre-formed 5-unsubstituted 4'-thio-pyrimidine nucleosides having protected or unprotected hydroxyl groups. When the hydroxyl groups on the 4-thio sugar compound are protected (for instance with ethers such as silyl ethers or esters such as acetate, benzoate or p-toluate esters), reaction with N-chlorosuccinimide in glacial acetic acid or with chlorine and iodobenzene and glacial acetic acid will introduce a 5-chloro substituent and reaction with iodine monochloride in dichloromethane will introduce a 5-iodo substituent, while reacting the unprotected 4'-thio sugar pyrimidine nucleoside with chlorine in carbon tetrachloride and acetic acid also introduces the 5-chloro substituent. Reaction with iodine and nitric acid also introduces the 5-iodo substituent. Reaction with bromine and acetic acid introduces a 5-bromo substituent to the unprotected nucleoside. Deprotection where necessary is by conventional techniques and is performed as the final step.

The 5-unsubstituted 4'-thionucleoside starting material of formula (II) (in which X^1 = H) may be produced as described above by coupling a 5-unsubstituted pyrimidine to a 4-thio sugar compound. Protection of the hydroxy groups of the 4-thio sugar moiety may be effected at any convenient stage.

X is C_{2-6} alkynyl

5-Alkynyl compounds may be produced by reacting a 5-iodo nucleoside of formula (II) wherein the hydroxyl groups of the 4-thio sugar are optionally protected (for instance by reaction of the unprotected nucleoside with p-toluoylchloride in pyridine to introduce p-toluoyl ester groups on the hydroxyl groups of the 4-thio sugar) with an appropriate alkynylating agent, for example trimethylsilyl acetylene or a terminal alkyne in the presence of a palladium catalyst such as bis(triphenylphosphine) palladium dichloride, and a copper catalyst such as cuprous iodide and triethylamine and, where necessary, removal of the protecting groups using sodium methoxide in methanol [c.f. M.J. Robins et al; Can. J. Chem., 60:554 (1982)].

Alternatively the 5-alkynyl group may be introduced by reacting a 5-iodo pyrimidine with trimethylsilyl-acetylene or a terminal alkyne in the presence of bis(triphenyl-phosphine)palladium dichloride, cuprous iodide, triethylamine and dimethylformamide followed, where necessary, by removal of the protecting groups and reacting the 5-alkynyl pyrimidine of formula (III) in suitably protected form (for instance the trimethylsilyl-protected form) with a protected 4-thio sugar compound as previously described followed by deprotection of the pyrimidine and sugar moieties as required.

X is C_{2-6} alkenyl

5-Alkenyl compounds may be produced by partial hydrogenation of the corresponding 5-alkynyl pyrimidine of formula (III) or of the nucleoside of formula (II) for instance using Lindlar catalyst poisoned with quinoline, and subsequently, in the case of the pyrimidine, coupling with a 4-thio sugar compound as described above.

Alternatively a 5-iodo nucleoside of formula (II) may be reacted with an appropriate alkenylating agent for example a 2-alkenoic acid ester (for instance the methyl ester) in the presence of palladium (II) acetate and triphenylphosphine to form the 5-(2-methoxycarbonyl alkenyl) derivative. The ester group is then removed by hydrolysis using sodium hydroxide forming the 2-carboxy alkenyl compound which itself is subjected to treatment with triethylamine in dimethylformamide at 100°C to give the 5-

vinyl analogue [c.f. S.G. Rahim et al., Nucleic Acids Research, 10(17):5285(1982)].

Yet another method for producing the 5-alkenyl compounds involves coupling the terminal alkene with a 5-iodo or 5-chloromercuri nucleoside of formula (II) (formed by for example reaction of the 5-unsubstituted nucleoside with mercury (II) acetate and sodium chloride), in the presence of a palladium catalyst such as palladium (II) acetate and a copper salt such as copper (I) chloride, or preferably a palladium catalyst such as dilithium palladium tetrachloride. Reaction of a 5-iodo- or 5-chloromercuri-nucleoside of formula (II) with allyl halides such as chloride or bromide in the presence of dilithium palladium tetrachloride leads to the formation of the corresponding 5-(alk-2-enyl) derivative which can be rearranged to form the 5-(alk-1-enyl) derivatives by treatment with tris(triphenylphosphine)rhodium chloride. This process may also be applied to the free pyrimidine base, which is subsequently condensed with the 4-thio sugar compound.

The above processes are exemplified by J.L. Ruth & D.E. Bergstrom, J. Org. Chem, 43 (14): 2870 (1978), J. Goodchild et al., J. Med. Chem, 26: (1983), D.E. Bergstrom & J.L. Ruth, J. Am. Chem. Soc., 98: 1587 (1976) and D.E. Bergstrom & M.K. Ogawa, J. Am. Chem. Soc., 100: 8106 (1978).

X is C_{2-6} haloalkenyl

5-(Haloalkenyl) substituents may be introduced into a nucleoside of formula (II) by conventional methods. For example, in order to prepare 5-(2-halovinyl) compounds the corresponding 5-(2-carboxyvinyl) nucleoside is treated with an appropriate halogenating agent, for example N-halosuccinimide in aqueous potassium acetate, or with potassium carbonate in dimethylformamide when the halogen is bromo or iodo. A 5-(2-chlorovinyl) nucleoside may also be made from the corresponding 5-(2-carboxyvinyl) nucleoside using chlorine gas in, for example, dimethylformamide (DMF).

Alternatively the 5-haloalkenyl group may be introduced into the appropriate free pyrimidine base to form a compound of formula (III) which is subsequently coupled with a 4-thio

compound as described above; this may be achieved for example by treating a 2,4-dimethoxy-protected 5-iodo-pyrimidine with an 2-alkenoic acid ester in the presence of palladium (II) acetate, triphenylphosphine and dioxane followed by removal of the methoxy protecting groups, hydrolysis of the ester with sodium hydroxide and reaction of the resulting 5-(2-carboxyvinyl) derivative with N-halosuccinimide (where halo is bromo or iodo) or chlorine gas (where halo is chloro) in the presence of a base such as sodium hydrogen carbonate in dimethylformamide. The 5-(2-carboxyvinyl) compound may also be produced by treating an unprotected 5-(hydroxymethyl)pyrimidine of formula (III) with an oxidising agent such as persulphate or manganese dioxide to form the corresponding aldehyde and followed by treatment of the aldehyde with malonic acid. The above processes are exemplifed by A.S. Jones et al, Tetrahedron Letts, 45; 4415 (1979) and P.J. Barr et al, J. Chem. Soc. Perkin Trans 1, 1981, 1665.

The 5-(2-haloalkenyl) base may alternatively be made by a novel route starting with a 2,4-dimethoxy protected 5-bromopyrimidine. This may be converted to the corresponding 5-lithium derivative by treatment with an organolithium reagent, preferably n-butyllithium at reduced temperature such as -70°C in an ethereal solvent such as diethylether. Reaction of the lithio derivative in situ with an appropriate ester of formic acid, such as ethyl formate at reduced temperature such as -70°C gives rise to the corresponding 5-formyl compound. Treatment of the formyl compound with malonic acid as described above gives rise to the 5-(2-carboxyvinyl) derivative. Similar halogenation gives rise to the required 5-(2-haloalkenyl) compound which is in the 2,4-dimethoxy protected from. Deprotection can then be carried out by conventional techniques.

5-Halovinyl compounds having more than one halogen substituent may be produced from a 5-halo-substitued 2,4-dimethoxy protected pyrimidine of formula (III) by reaction with a strong base such as butyl lithium and the resulting lithio derivative treated with the appropriate haloalkene followed by removal of the protecting groups and coupling to the 4-thio sugar compound as described above [c.f. P.L. Coe et al., J. Med. Chem. 25:1329 (1982)].

Alternatively, the halogen atoms may be introduced sequentially into a 5-substituent of the pyrimidine base. Thus, for example treatment of 5-acetyl uracil with a chlorinating agent such as phosphorus oxychloride provides the 5-(1-chlorovinyl) group with simultaneous chlorination of the hydroxyl groups of the pyrimidine base. Treatment with potassium ethoxide then hydrogen chloride and finally bromine leads to bromination of the 5-unsaturated side chain of the pyrimidine base with simultaneous conversion of the 2,4-dichloro groups on the pyrimidine ring to form the corresponding uracil derivative. The resulting pyrimidine base can then be coupled to the 4-thio sugar compound as described above [c.f. P.J. Barr et al. Nucleic Acids Res. 3: 2845 (1976) and P.J. Barr et al., J. Chem. Soc. Perkin Trans 1, 1981: 1665].

X is C_{2-6} alkyl

5-C_{2-6} Alkyl eg. 5-ethyl substituted nucleosides may be produced by hydrogenation of the corresponding 5-alkynyl or 5-alkenyl pyrimidine base followed by coupling to the 4-thio sugar compound. Conventional hydrogenation conditions, such as hydrogen over palladium/charcoal catalysts, may be adopted.

X is trifluoromethyl

5-Trifluoromethyl uracil is commercially available and this may be condensed with a 4-thio sugar compound in accordance with process B described above. The 5-trifluoromethyl cytosine analogue may be made from the uracil compound/using an analogous procedure to that described by Sung as mentioned below.

X is 5-haloalkyl

5-Fluoroalkyl substituents may be generated from the corresponding 5-hydroxyalkyl substituents, preferably starting from nucleosides having protected sugar hydroxyl groups on the 4-thio sugar moiety. Suitable protecting groups include tert-butyl diphenylsilyloxy groups which may be introduced using tert-butyldiphenylsilylchloride. The protected 5-hydroxyalkyl nucleoside is treated with a fluorinating agent such as diethylaminosulphurtrifluoride followed by deprotection of the

hydroxyl groups using tetra-n-butylammonium fluoride to give the monofluoroalkyl derivative. Alternatively treatment of the 4'-thio sugar- protected 5-hydroxyalkyl nucleoside with manganese dioxide or pyridinium dichromate produces the corresponding aldehyde which may be treated with diethylamino- sulphur trifluoride. Treatment with tetra-n-butylammonium fluoride removes the protecting groups and liberates the 5-difluoroalkyl derivative.

Other 5-haloalkyl, for example haloethyl, substituents may also be generated using the corresponding 5-hydroxyalkyl substituents. The 5-hydroxyalkyl compound, in the form of either a base or a nucleoside is reacted with carbon tetrachloride and triphenylphosphate to introduce a chloro substituent, or with N-bromosuccinimide and triphenylphosphate to introduce a bromo substituent, or with N-bromosuccinimide, triphenylphosphine and tetrabutyl-ammonium iodide to introduce an iodo substituent [c.f. J.D. Fissekis & F. Sweet, J. Org. Chem. 1973, 38, 264, and WO84/00759].

The above 5-hydroxyalkyl nucleoside starting materials where the alkyl group is a methylene are obtained from the corresponding 5-methyl-nucleosides by protection (for instance using tert-butyldiphenylsilylchloride) of the hydroxyl groups of the 4-thio sugar moiety, photolytic bromination (for instance, using bromine, N-bromosuccinimide in carbon tetrachloride) and hydrolysis of the bromoalkyl side chain using sodium bicarbonate.

X is nitro or optionally substituted amino

Nitro-substituents are introduced at the 5-position of the 5-unsubstituted 4'thio-nucleosides by reaction with a nitrating agent for example nitronium tetrafluoroborate (NO_2BF_4), and these may be reduced using hydrogen over palladium/charcoal or tin (II) chloride to provide the corresponding amino substituent. [c.f. G-F. Huang and P.F. Torrence, J. Org. Chem., 42: 3821 (1977)]. 5-Nitro-substituted pyrimidines are readily available and may be coupled with 4-thio sugar compounds as described above.

5-Alkylamino and 5-dialkylamino substituents may be introduced by reacting a suitably protected 5-iodo-nucleoside with a corresponding alkylamine or dialkylamine. Protection is

preferably by acylation for example by acetylation using acetic anhydride in pyridine.

X is alkoxy

Alkoxy substituents are introduced at the 5-position by reaction of the corresponding 5-hydroxy-4'-thio-nucleoside with a base, for example sodium hydroxide followed by alkylation with an appropriate alkyl halide in a suitable solvent such as methanol. The starting 5-hydroxy 4'-thio-nucleoside may be obtained from the 5-unsubstituted 4'-thio-nucleoside by treatment with bromine in an aqueous solvent such as aqueous tetrahydrofuran followed by treatment with base such as trimethylamine.

Alternatively, alkoxy substituents may be introduced by treatment of the corresponding 5-iodo-4'-thionucleoside with an alkoxylating agent such as sodium alkoxide in an appropriate solvent such as methanol or dimethylformamide or the corresponding alkanol.

X is cyano

Cyano substituents are introduced at the 5-position by reaction of the corresponding 5-iodo 4'-thio-nucleoside with potassium cyanide in the presence of potassium acetate in a suitable solvent such as dimethylformamide, preferably at elevated temperature, for example 80°C-120°C, preferably 100°C [c.f. P.F. Torrence & B. Bhoosham J. Med. Chem., 20, 974 (1977)].

X is thiocyanate, alkylthio, mercapto

Compounds of the formula (I) with these substituents may be prepared from a suitably protected 5-halogen-nucleoside (for example bromo or iodo) by methods analogous to those described in Lin et al. J Med Chem, 31, 336-340 1988. Methods for the synthesis of a 5-halogen-nucleoside are described in the above mentioned specifications.

X is hydroxy

5-hydroxy-4'-thio-nucleosides may be prepared by the method described above in connection with the preparation of alkoxy

compounds. The starting compound, 5-unsubstituted 4'-thio-nucleoside may conveniently be prepared by condensation of the appropriate sugar moiety with commercially available uracil.

X is hydroxy-C_{1-3}alkyl.

These compounds may be prepared as described above in connection with the preparation of haloalkyl compounds. Hydroxymethyl uracil itself is commercially available.

X is C_{1-6}alkoxyC_{1-2} alkyl or C_{1-6}alkylthiomethyl.

Compounds in which X is alkoxymethyl or alkylthiomethyl may be made starting from bases in which the group X is of the formula $-CH_2OH$. The alkoxymethyl compounds may be made by reacting this starting material with an appropriate alkanol group in the presence of an acid catalyst or an acidic ion exchange resin. The alkylthiomethyl compounds may be made in a similar way but using the appropriate alkylmercaptan group or an appropriate metal salt thereof. The resulting base may be condensed with the desired 4-thio sugar as described herein.

The corresponding alkoxyethyl compounds may be made in an analogous manner starting from the appropriate base in which the group X is hydroxyethyl. These starting bases are either commercially available or may be made as described above in connection with the preparation of haloalkyl compounds.

Alternatively these alkoxyalkyl and alkylthiomethyl compounds may be made from nucleosides of the formula I or a protected derivate thereof in which the group X is $-CH_2L$ where L is a leaving group, eg halo such as bromo, or alkyl or arylsulphonyloxy such as trifluoromethanesulphonyl or p-toluenesulphonyl or a secondary acyclic or cyclic amino group, such as dimethylamino or pyrrolidinyl. The reaction is carried out by treatment of one of these with a suitable nucleophilic reagent serving to introduce the O-alkyl or S-alkyl group, eg. alkyl-OH, alkyl-SH or alkyl-SM where M is a metal ion such as sodium. The procedure may be performed using methods analogous to those described by Barwolff and Langen, Nucleic Acid Chemistry - Improved and New Synthetic Procedures, Methods and Techniques, Part 1, Eds. L.B. Townsend and R.S. Tipson, p359.

The above reference also describes the procedures which may be utilised to make compounds in which L is OH. Such compounds may be used to make compound where L is O-alkyl or S-alkyl using the procedures described above.

The compounds where L is dimethylamino or pyrrolidinyl may be prepared by methods analogous to those described by Badman and Reese in J. Chem. Soc. Commun. 1987, 1732-1734 and by Jones et al, Synthesis 1982, 259-260.

X is formyl

Compounds of formula I wherein X is formyl are typically prepared from the corresponding compound wherein X is CH_2L, and L is Br. The latter compound may be prepared by methods analogous to those described by Barwolff and Langen, Nucleic Acid Chemistry - Improved and New Synthetic Procedures, Methods and Techniques, Part 1, Eds. L.B. Townsend and R.S. Tipson, p359.

Sugar moieties

The 4-thio-sugar compound may be produced by conventional methods prior to coupling with the base or derived by modification of another sugar moiety which is already part of a nucleoside. When the compounds of formula I are produced in accordance with process (C), the process may be carried out using the following procedures to prepare compounds of formula (I) in which R^2 and R^3 have the following meanings include:-

a) R^2 is hydrogen and R^3 is hydroxy.

The synthesis of the 2'-deoxy sugar moiety may be conducted in accordance with the methods disclosed in the examples which follow. Reference may also be made to the methods described in M.J. Robins, T.A.Khwaja, R.K. Robins, J. Org. Chem., 1970, 35(3) 636.

b) R^2 and R^3 together form a carbon-carbon bond.

Such compounds may be prepared from a corresponding 3'5',-anhydro compound for example by treatment with a strong base eg. potassium tert-butoxide. Such 3',5'-anhydro compounds may be

prepared by treating the corresponding 3',5'-methanesulphonate diester with a base. The 3'5'-methanesulphonate diester may be obtained by esterification of the 2-deoxy-L-ribose sugar which may be synthesised by analogous methods to those of Smejkal and Sorm, (1964), Nucleic Acids. Components and their Analogues, part Lii, volume 29, 809.; Genu-Dellac et al., Nucleosides and Nucleotides, 10 (6), 1345-1376, (1991); Robins et al. J. Org. Chem. 35 (3), 636-639 (1970),; and Schimmel & Bevill, Analytical Biochemistry 37, 385-394, (1970).

c) R^2 and R^3 are both hydrogen.

The 2,3-dideoxy-4-thio-L-ribonucleosides may be obtained by a method analogous to that disclosed by E.J. Prisbe and J.C. Martin (Synth. Commun. (1985), 15(4), 401-409), which describes the synthesis of D-dideoxy nucleosides from 2-deoxy nucleosides, or by a method analogous to J.A. Secrist et al (J. Med. Chem. (1992) 35,533-539) which discloses the synthesis of D-dideoxy-4-thionucleosides from L-glutamic acid. The use of D-glutamic acid (Aldrich Chemical Company Ltd) will result in the synthesis of dideoxy-4-thio-L-nucleosides.

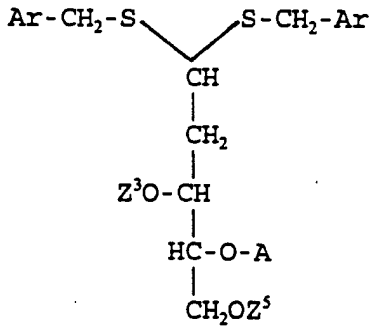
The above reactions are all suitable for producing uracil nucleosides; most of such reactions may also be used to form cytosine nucleosides. When this is not convenient or possible, cytosine analogues can be prepared most conveniently from the uracil compounds using an analogous procedure to that described by W.L. Sung, J. Chem. Soc. Chem. Commum., 1981, 1089]: for example the acetylated uracil nucleoside (produced for instance by reactions as described above and acetylated using acetic anhydride in pyridine) is treated with p-chlorophenyl-phosphorodichloridate, 1,2,4-triazole and pyridine to produce the 4-(1,2,4-triazol-1-yl) derivative which is then treated with ammonia in dioxane (which also removes the 4-thio sugar protecting group(s)) to form the corresponding unprotected cytosine 4'-thionucleoside.

The derivatives of the compounds of formula (I) may be prepared in conventional manner. For example, esters may be prepared by treating a compound of formula (I) with an appropriate esterifying agent, for example, an acyl halide or

anhydride. Salts may be prepared by treating a compound of formula (I) with an appropriate base, for example an alkali metal, alkaline earth metal or ammonium hydroxide, or where necessary, an appropirate acid, such as hydrochloric acid or an acetate, eg. sodium acetate.

The anomers of compounds of the formula (I) may be separated by conventional means, for example by chromatography or fractional crystallisation.

In a further aspect of the invention, compounds of the formula (V) in which in which R^2 is hydrogen, R^{3a} is OZ^3 and W is a group -S-CH_2-Ar as defined above may be made by ring closure of a compound of the formula (VII)



(VII)

where Z^3 and Z^5 are hydroxyl protecting groups as defined above, for example optionally substituted benzyl or acyl groups as defined above. Preferably the groups Z^3 and Z^5 are acyl groups. The group A is a leaving group, for example an organosulphonyl group such as an optionally substituted alkyl- or aryl-sulphonyl group, for instance methanesulphonyl, a haloalkylsulphonyl group (eg. trifluoromethylsulphonyl) and optionally substituted phenyl-sulphonyl (eg. toluylsulphonyl or bromobenzenesulphonyl), and Ar is as defined above. A is preferably a methanesulphonyl group.

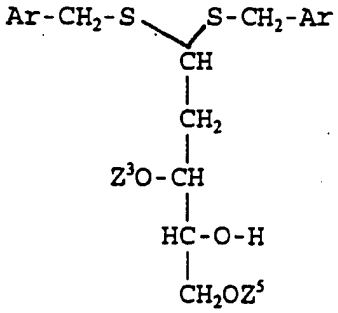
The ring closure may be performed under appropriate basic conditions. Suitable conditions include those described by J. Harness and N.A. Hughes (Chem. Comm. 1971, 811), which includes the use of sodium iodide and barium carbonate.

Preferably the reaction is carried out in a solvent such as acetone or dimethylformamide (DMF). DMF is preferred. The use of sodium iodide to debenzylate the intermediate arylthio cation is also preferred. Triethylamine may also be used as an

alternative base.

The compound of the formula (V) in which R^2 is hydrogen, R^{3a} is OZ^3 and W is $-S-CH_2-Ar$ may be converted to a compound of formula (V) where W represents other leaving groups by techniques known in the art, for example those disclosed in EP-A-0 409 575. For example, the compounds may be reacted with an appropriate acylating agent such as acetic anhydride (optionally in the presence of acetic acid), in the presence of a mineral acid such as sulphuric acid. This will provide compounds in which the group W is acyloxy, which is a suitable leaving group for the reactions described herein. The acyloxy group may be converted to other leaving groups, eg. halo groups, using known methods.

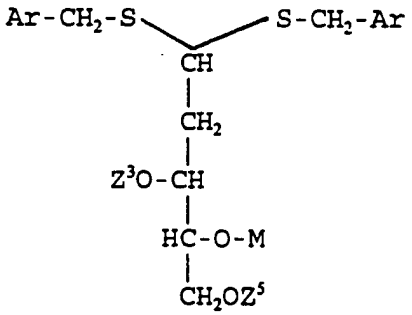
The compound of the formula (VII) may be made from a compound of formula (VIII)



(VIII)

where Ar, Z^3 and Z^5 are as defined above. Preferably the groups Z^3 and Z^5 are acyl groups. Conversion of a compound of formula (VIII) to a compound of formula (VII) is carried out according to standard procedures such as treatment with an appropriate optionally substituted alkyl- or aryl-sulphonyl halide, eg. methanesulphonylchloride in a basic solvent such as pyridine. Other suitable alkyl or aryl sulphonyl halides include trifluoro-methanesulphonyl chloride and p-toluene-sulphonyl chloride.

The compound of formula (VIII) may be made from a compound of formula (IX):

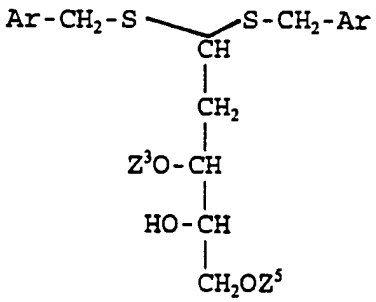


(IX)

where Ar, Z^3 and Z^5 are as defined above and M is a hydroxyl protecting group which may be removed under conditions which leave the -S-CH_2-Ar groups and the groups Z^3 and Z^5 in place.

Preferably, the group M is a group of the formula Ar^1-CO- where Ar^1 is a phenyl group which may be optionally substituted by any of the substituents described above for the group Ar. Removal of this group M may be performed under standard conditions, for example with a base such as an alkali metal alkoxide, for instance sodium methoxide in methanol.

The compounds of formula (IX) may be obtained by the concomitant inversion and derivatization of the 4-hydroxy group of a compound of formula (X):

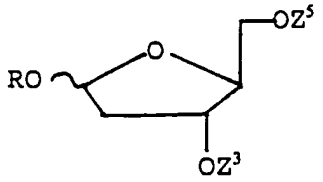


(X)

wherein Ar, Z^3 and Z^5 are as defined above. The inversion and derivatization may be effected by reacting the compound of formula (X) with a derivative of the group M, such as an acid of the formula Ar^1-COOH, for example benzoic acid (or a reactive derivative thereof) where Ar^1 is as defined above. The reaction is performed typically at room temperature and under neutral conditions in a suitable polar solvent, for instance tetrahydrofuran. Preferably the Mitsunobu reaction is used for

the inversion and derivatization; diethyl azodicarboxylate (DEAD) and triphenylphosphine are used as coreactants together with the acid Ar^1COOH.

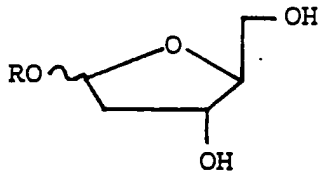
The compound of formula (X) may be made from a glycoside compound of formula (XI)



(XI)

where Z^3 and Z^5 are as defined above and R is a hydrocarbyl group such as a C_{1-4} hydrocarbyl group, eg. a C_{1-4} alkyl group, preferably methyl. The compound of the formula (X) is reacted under acid conditions at an elevated temperature with a compound of formula $Ar-CH_2-SH$, where Ar is as defined above. Suitably the reaction is performed in the presence of hydrochloric acid which may be in aqueous or anhydrous form. Preferably the elevated temperature is from 30°C to 60°C, for example 40°C. When Ar is a phenyl group, the compound $Ar-CH_2-SH$ will be benzyl thiol.

Compounds of the formula (XI) may be made from a compound of formula (XII)



(XII)

where R is a defined above. The hydroxyl groups of the compound of formula (XII) are protected under conventional conditions with the reactive derivative of the groups Z^3 and Z^5. Suitably, the bromo derivative may be used. Thus when Z^3 and Z^5 are benzyl groups, benzyl bromide may be used. The reaction may be performed in an organic solvent such as tetrahydrofuran in the presence of a suitable base such as sodium hydride and a phase transfer catalyst such as tetrabutylammonium iodide.

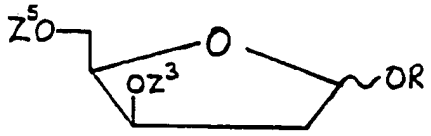
Compounds of the formula (XII) may be made by standard techniques from 2-Deoxy-L-ribose, which can be made by methods

described in M.J. Robins, T.A.Khwaja, R.K. Robins, J. Org. Chem., 1970, 35(3) 636. 2-Deoxy-L-ribose may be reacted with an alcohol of formula R-OH (where R is as defined above) in the presence of an acid. Hydrochloric acid is suitable. When R is a methyl group, the alcohol R-OH will be methanol.

The conversion of 2-deoxy-L-ribose to a compound of formula (XII) will also produce a small proportion of the corresponding pyranoside compound, substituted at the 1-position by the group -OR. This may remain in the reaction mixture during the converions of (XI) to (X), (X) to (IX) and the subsequent reactions described above and it will undergo analogous reactions. These by-products may be separated at any convenient step by conventional means, eg. chromatography.

The compound of the formula (VIII) may also be made directly from the compound of formula (X) using a Mitsunobu reaction under conditions analogous to those described by D.R. Williams et al, JACS (1990) 112, 4552.

Compounds of the formula (VIII) may also be made by reaction of a compound of the formula (XIII)



(XIII)

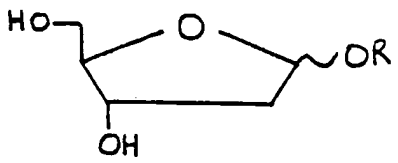
where R, Z^3 and Z^5 are as defined for a compound of formula (XI); with a compound of the formula Ar-CH_2-SH where Ar is defined above. The reaction may be conducted using similar conditions to those described above for the preparation of the compound of the formula (X). The reaction is performed in the presence of an acid, for example an inorganic acid such as HCl or a Lewis acid such as $TiCl_4$. $TiCl_4$ is preferred.

The groups Z^3 and Z^5 are preferably acyl groups, in particular p-nitrobenzoyl groups.

A compound of the formula Ar-CH_2-SH which is preferred include p-methoxybenzyl mercaptan. When compounds of the formula (I) are made using a synthetic route which includes the production of compounds of formula (VIII) from compounds of formula (XIII), this substituent is preferred, desirably in

conjunction with the groups Z^3 and Z^5 being p-nitrobenzoyl.

Compounds of the formula (XIII) may be prepared from compound of the formula (XIV)



(XIV)

where R is as defined above, using a Mitsunobu reaction to protect the 3′ and 5′ hydroxy groups and to invert the 3-hydroxyl of the ribo-sugar ring at the same time. The compound of formula (XIV) is reacted with a compound or compounds of formula Z^nOH where Z^n is Z^3 and/or Z^5.

Desirably Z^3 and Z^5 will be the same and a single compound Z^nOH may be used. If different values of Z^3 and Z^5 are required, then the required mixture of compounds of Z^nOH may be used, and the desired reaction products separated from the resulting reaction mixture.

Preferred compounds of the formula Z^nOH are those where Z^n is an acyl group as defined above. Preferably, Z^nOH is p-nitrobenzoic acid, although other benzoic acids may also be used.

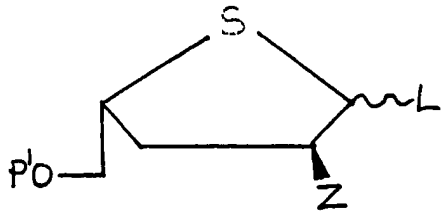
The reaction is performed in the presence of an azidocarboxylate such as diethylazidodicarboxylate or preferably diisopropylazidodicarboxylate. The solvent for the reaction may be DMF, tetrahydrofuran, dichloromethane or toluene. Toluene is preferred. The reaction may be performed at room temperature.

When compounds of the formula (I) are made starting from compounds of the formula (XIV), it is preferred that Z^3 and Z^5 are both p-nitrobenzyl.

Compounds of the formula (XIV) may be made from 2-deoxyribose using techniques known in the art, for example as described above in connection with the production of compounds of formula (XII).

Compounds of the formula (I) may also be made by reaction

of a compound of formula (III) with a compound of formula (XV)



(XV)

where L is a leaving group, for example, an acyloxy group such as C_{1-4} alkanoyloxy, for instance, acetoxy; P^1 is a protecting group or hydrogen, and Z is a directing group.

Suitable groups P^1 include groups such that P^1O is an ether group, e.g. a silyl ether group (such as tertbutyldiphenylsilyl ether or tertbutyldimethylsilyl ether), a straight or branch chain alkyl ether group, a cyclic ether group (such as tetrahydropyran-2-yl ether) or an optionally substituted aryl ether group (such as benzyl ether, trityl ether or benzhydryl ether). The group P^1O- can also be an ester group e.g. wherein P^1 is QC=O where Q is alkyl (such as methyl), cycloalkyl or an optionally substituted aryl.

Suitable groups Z include sulphenyl groups and selenyl groups. The preferred group Z is phenylselenyl.

The reaction of the compound of formula (XV) with the base of formula (III) may be carried out for example in the presence of nonafluorobutane sulphonic acid or a Lewis acid catalyst, e.g. tin (IV) chloride, a mercury (II) salt or trimethylsilyl triflate. The reaction can be carried out, for example, at a temperature of from 0°C to room temperature in a suitable solvent such as acetonitrile or a chloroalkane.

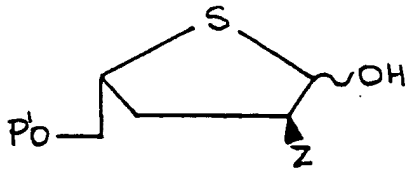
Following reaction of a compound of formula (XV) with a compound of formula (III), the group Z may be eliminated to provide a compound of formula (I) in which R^2 and R^3 together form a carbon-carbon bond. The protecting group P^1 may be removed either before or after elimination of Z.

For example when Z is phenylselenyl it may be eliminated under oxidising conditions which are capable of oxidising selenium without oxidising sulphur, e.g. by treatment with m-chloroperbenzoic acid in dichloromethane at -20°C (Toru *et al*, Tetrahedron Letters, 1986, 27; 1583).

Compounds of the formula (I) in which R^2 and R^3 are both hydrogen may be made by elimination of the group Z (from the reaction product of (XV) and (III)) under reducing conditions, e.g. using tributyltin hydride and triethyl borane (see Nozaki *et al*, Tetrahedron Letters, 1988, 29; 6125).

Reference may also be made to EP-A-514 036 which describes the production of D-thionucleosides from, *inter-alia*, the isomer of the compound of formula (XV) which has the D-configuration. The reactions described in EP-A-514 036, the contents of which are incorporated herein by reference, may be applied to the production of compounds of formula (I).

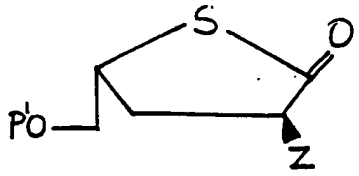
Compounds of the formula (XV) may be made by acylation under standard conditions (eg. acetic anhydride with pyridine) of a compound of formula (XVI):



(XVI)

in which P^1 and Z are as defined above.

The compound of formula (XVI) may be made from a compound of formula (XVII),



(XVII)

in which P^1 and 2 are as defined above, by reduction of the carbonyl group of (XV), for example using conventional reagents such as diisobutylaluminium hydride.

Compounds of the formula (XVII) may be prepared by methods analogous to those described in EP-A-514 036 but starting from R-(+)-glycidol instead of the (S)-(-)-enatiomer described in this reference. Example 13 also describes the preparation of 4-(R)-(tert-butyl-diphenylsiloxy-methyl)-2-(S)-phenylselenyl-4-thio-butanoluctone, and those of skill in the art may refer to Example 13 for the preparation, by analogous methods, of other compounds of the formula (XVII).

The anomerisation of α- or β-anomers of compounds of the formula (I) into β- or α-anomers respectively may be perfomed using known techniques, see for example Yamaguchi, T. and Saneyoshi, M., Chem. Pharm. Bull. 32, 1441-1450 (1984) vol. 4. The process may also be conducted by the use of an anhydride in the presence of a strong acid, as disclosed in British Patent application 9218737.6, in the name of the University of Birmingham, filed on 4th September 1992.

The invention is illustrated by the following Examples.

Example A

Preparation of Methyl 3,5-di-O-benzyl-2-deoxy-L-erythro-pentoside

To a solution of 2-deoxy-L-ribose (50g, 373 mmol) in dry methanol (900ml) is added a 1% solution of dry hydrogen chloride in methanol (100ml). The mixture is kept in a stoppered flask for 30 minutes after which the reaction is stopped by adding, with vigorous stirring, silver carbonate (10g). The mixture is filtered by gravity and the colourless filtrate evaporated to a syrup using a dry rotary evaporator. Residual methanol is then removed by repeated evaporation with dry THF. The syrup is then dissolved in dry THF (470ml). Under an atmosphere of dry nitrogen, at 0°C, with stirring sodium hydride in a 50% oil-dispersion (39.4g, 821 mmol) is slowly added to the THF mixture. Next, dry tetrabutylammonium iodide (30.3g, 82.1 mmol) is added followed by benzyl bromide (140g, 821 mmol), which is added over 1 hour. The THF is removed in vacuo, the residue dissolved in dichloromethane and then poured into ice/water. The dichloromethane solution is extracted from this mixture and then dried over magnesium sulphate. The dichloromethane is evaporated under reduced pressure and the resulting residue applied to a silica gel column eluted with hexane-ethyl acetate (4:1). Combination of the appropriate fractions gives the α and ß isomers of the title product as a syrup.

Preparation of 3,5-di-O-benzyl-2-deoxy-L-erythro-pentose

dibenzyl dithioacetal

Concentrated hydrochloric acid (150ml) is added dropwise to a stirred mixture of methyl 3,5-di-O-benzyl-2-deoxy L-erythro-pentoside (77.5g, 236 mmol) and benzyl thiol (147g, 1.19 mol) at room temperature. The temperature is then raised to 40°C and the mixture stirred for 18 hours. The mixture is dissolved in chloroform, poured into ice/water, neutralised with sodium hydrogen carbonate and extracted with chloroform. The chloroform extracts are dried over magnesium sulphate and the chloroform evaporated under reduced pressure. The residue is applied to a silica gel column and eluted with hexane-ethyl acetate (4:1) to give the title product.

Preparation of 4-O-benzoyl-3,5-di-O-benzyl-2-deoxy-D-threo-pentose dibenzyl dithioacetal

To a solution of 3,5-di-O-benzyl-2-deoxy-L-erythro-pentose dibenzyl dithioacetal (54.1g,99.3 mmol),triphenyl-phosphine (39.1g, 149 mmol) and benzoic acid (18.2g, 149 mmol) in dry THF (800ml) is added a solution of DEAD (26.0g, 149 mmol) in dry THF (200ml) dropwise, with stirring, at room temperature for 18 hours. The THF is removed in vacuo and the residue applied to a silica gel column eluted with hexane-ethyl acetate (85:15). Combination of the appropriate fractions gives the title product.

Preparation of 3,5-di-O-benzyl-2-deoxy-D-threo-pentose dibenzyl dithioacetal

To a solution of 4-O-benzoyl-3,5-di-O-benzyl-2-deoxy-D-threo-pentose dibenzyl dithioacetal (88.8g, 137 mmol) in dichloromethane (500ml) is added a solution of sodium methoxide (11.1g, 206 mmol) in methanol (205ml) dropwise, with stirring, at 0°C. The reaction mixture is allowed to warm to room temperature over a period of 3 hours. The mixture is then poured into a 5% solution of NaH_2PO_4 and extracted with dichloromethane. The dichloromethane extracts are then washed with a 5% solution of sodium hydrogen carbonate and water, dried (magnesium sulphate) and evaporated. The crude title product is applied to a silica gel column eluted with hexane-ethyl acetate

(4:1). Combination of the appropriate fractions gives the title product.

Preparation of 3,5-di-O-benzyl-2-deoxy-4-O-methane-sulphonyl-D-threo-pentose dibenzyl dithioacetal

To a solution of 3,5-di-O-benzyl-2-deoxy-D-threo-pentose dibenzyl dithioacetal (61.4g, 113 mmol) in dry pyridine (700ml) is added methanesulphonyl chloride (19.4g, 169 mmol) in dry pyridine (200ml) dropwise, with stirring, at 0°C. The temperature of the mixture is raised to room temperature and stirring continues for 18 hours. The pyridine is then removed in vacuo and the residue dissolved in dichloromethane. The dichloromethane extracts are then successively washed with 2M hydrochloric acid, 1M sodium carbonate and water, dried (magnesium sulphate) and evaporated to give the title product.

Preparation of benzyl 3,5-di-O-benzyl-2-deoxy-1,4-dithio-L-erythro-pentofuranoside

A suspension of 3,5-di-O-benzyl-2-deoxy-4-O-methanesulphonyl D-threo-pentose dibenzyl dithioacetal (29.4g, 47.4 mmol), sodium iodide (74.0g, 494 mmol), barium carbonate (148g, 750 mmol) and dry acetone (1L) is boiled under reflux for 42 hours. At the end of this time the suspension is filtered and the solids washed with chloroform. The filtrate is sequentially washed with water, sodium thiosulphate solution (5%) and water, dried (magnesium sulphate) and evaporated. The resultant residue is applied to a silica gel column, eluted with hexane-ethyl acetate (9:1). Combination of the appropriate fractions gives the title product.

Preparation of 3',5'-di-O-benzyl-4'-thio-L-thymidine and its α-anomer

A suspension of benzyl 3,5-di-O-benzyl-2-deoxy-1,4-dithio-L-erythro-pentofuranoside (22.5g, 51.6 mmol), bis TMS-thymine (46g, 170 mmol), mercuric bromide (20.5g, 56.7 mmol), cadmium carbonate (29.3g, 170 mmol) and dry toluene (1L) is boiled under

reflux, with stiring, for 24 hours. The hot mixture is then filtered and the solids are washed with toluene. The filtrate is successively washed with potassium iodide solution (30%) and water and then evaporated. The residue is taken up in 4:1 methanol-water, stirred for 30 minutes, the suspension filtered and the filtrate evaporated. The residue is applied to a silica gel column (hexane-ethyl acetate (1:1)) and combination of the appropriate fractions gives the title product.

Preparation of ß-4'-thio-L-thymidine

To a 2M boron trichloride solution in dry dichloromethane (55ml) cooled to -78°C, is added a solution of ß-3',5'-di-O-benzyl-4'-thio-L-thymidine (1.6g, 3.7mmol) in dry dichloromethane (30ml). Stirring is continued for 5 hours at -78°C. This is followed by the dropwise addition of a 1:1 methanol-dichloromethane solution (200ml) over 40 minutes. The reaction mixture is allowed to warm to room temperature over 1 hour and the solvent removed in vacuo and coevaporated with dry methanol (3 x 30 ml). The residue is applied to a silica gel column eluted with chloroform-methanol (85:15) to give the title product.

Preparation of benzyl 2-deoxy-1,4-dithio-3,5-di-O-p-toluoyl-L-erythro-pentofuranoside

To a 2M boron trichloride solution in dry dichloromethane (150ml) cooled to -78°C, is added a solution of benzyl 3,5-di-O-benzyl-2-deoxy-1,4-dithio-L-erythro -pentofuranoside (4.2g, 10mmol) in dry dichlormethane (100ml), dropwise, over 30 minutes. Stirring is continued for 5 hours at -78°C. This is followed by the dropwise addition of a 1:1 methanol-dichloromethane solution (200ml) over 40 minutes. The reaction mixture is allowed to warm to room temperature over 1 hour and the solvent removed in vacuo and coevaporated with dry methanol (3x30ml). The crude residue is dissolved in dry pyridine (25ml), cooled to 0°C, and a solution of p-toluoyl chloride (4.6g, 30 mmol) in dry pyridine (25ml) added, dropwise, with stirring. The pyridine is removed in vacuo, the residue extracted with chloroform, and the extract successively washed with 2M hydrochloric acid, 1M sodium

carbonate and water, dried (magnesium sulphate) and evaporated. The residue is applied to a silica gel column eluted with hexane-ethyl acetate (9:1) to give the title product.

Preparation of E-5(2-bromovinyl-2'-deoxy-4'-thio-3',5'di-O-p-toluoyl-L-uridine and its α-anomer

To a solution of benzyl 2-deoxy-1,4-dithio-3,5-di-O-p-toluoyl-L-erythro-pentofuranoside (1.4g,2.8 mmol) in carbon tetrachloride (15ml) is added a solution of bromine (0.49g, 3.1mmol) in carbon tetrachloride (15ml) with stirring at room temperature. After 5 minutes the mixture is concentrated under diminished pressure and then carbon tetrachloride (5ml) added and the mixture evaporated to remove the excess bromine. The evaporation procedure is repeated four times. The resulting syrupy bromide is used directly in the next step.

To a solution of the bromide in carbon tetrachloride (10ml) is added the bis TMS-derivative of E-5-(2-bromovinyl)uracil (1.7g, 4.7mmol) in carbon tetrachloride (10ml). The mixture is stirred until homogenous, evaporated and the residue heated for 1 hour at 90-100°C. The cooled, dark residue is dissolved in 4:1 methanol-water (30ml), the solution boiled for 15 minutes under reflux and then evaporated. The residue is triturated with chloroform (40ml) and the solid 5-(2-bromovinyl) uracil that separates filtered off. The filtrate is successively washed with aqueous sodium hydrogen carbonate and water, dried (sodium sulphate) and evaporated. The residue is applied to a silica gel column eluted with hexane-ethyl acetate (3:2). Combination of the apporpirate fractions gives the title product.

EXAMPLE 1

Preparation of E-5-(2-bromovinyl)-2'-deoxy-4'-thio-β-L-uridine.

E-5-(2-bromovinyl)-2'-deoxy-4'-thio-3,'5'-di-O-p-toluoyl-β-uridine (200mg, 0.34mmol) is dissolved in a solution of sodium methoxide in methanol (7.5ml,0.1m) and the mixture is allowed to stand at 22°C for 24 hours. The solution is neutralised by careful addition of Dowex 50 ion exchange resin (H^+form) to pH6. The resin is filtered off and washed with methanol and the filtrate and washings evaporated to a white solid. This is

applied to a silica gel column eluted with chloroform-methanol (9:1). Combination of the appropriate fractions gives E-5-(2-bromovinyl)-2'-deoxy-4'thio-β-L-uridine which is crystallised from methanol-water.

EXAMPLE B

3',5'-Di-O-benzyl-2'-deoxy-5-iodo-4'-thio-L-uridine

Mercuric bromide (370 mg; 1.03 mmol) and cadmium carbonate (480 mg; 2.8 mmol) are added to a stirred solution, protected from moisture, of benzyl 3,5-di-O-benzyl-2-deoxy-1,4-dithio-L-erythro-pentofuranoside (436 mg; 1.0 mmol) in dry MeCN (3 ml). A solution of 5-iodo-bis-O-trimethylsilyluracil (3 mmol) in MeCN (12 ml) is added via syringe. The progress of the reaction is monitored by analytical HPLC while the mixture is heated under reflux for 1 h. When cooled to ambient temperature, water (200 µl) is added and after stirring for 30 min. the suspension is filtered. The filtrate is evaporated and redissolved in dry MeCN, the precipitated 5-iodouracil is removed by filtration. The filtrate is purified by preparative HPLC on a 2.5 cm (1 in.) Zorbax C8 reverse phase column eluted at 20 ml min^{-1} with a gradient [0 - 95% MeCN-water containing a constant 0.2% trifluoroacetic acid] over 20 min.; half-minute fractions are collected. Fractions containing pure product are pooled.

EXAMPLE 2

2'-Deoxy-5-iodo-4'-thio-L-uridine

The above product (240 mg; 0.44 mmol) dissolved in dry CH_2Cl_2 (10 ml + 2 ml rinse) is added over 30min to a stirred 1M solution of BCl_3 in CH_2Cl_2 (18 ml, 18 mmol) at -78°C under N_2. The reaction is followed by analytical HPLC. After 6h at -78°C, MeOH-CH_2Cl_2 (1:1, 18 ml) is added slowly and the mixture allowed to warm to ambient temperature then evaporated. The residue is re-evaporated from MeOH (3x), taken up in MeOH-$CHCl_3$ (1:1, 15ml) and the solid collected by filtration, yielding the desired ß-anomer of the product.

EXAMPLE 3

2'-Deoxy-5-ethyl-4'thio-L-uridine

Benzyl 3,5-di-O-benzyl-2-deoxy-1,4-dithio-D-erythro-pentofuranoside (5.6 mmol) is dissolved in CCl_4 (30 ml) and bromine (6.2 mmol) in CCl_4 (30 ml) is added. After stirring for 5 min. at ambient temperature the solvent is evaporated and the residue re-evaporated from CCl_4 (10 ml) to remove excess bromine. To a solution of this crude 1-bromo-thiosugar in CCl_4 (15 ml) is added bis-O-trimethylsilyl-5-ethyl uracil (16.6 mmol) [prepared by refluxing 5-ethyluracil (16.6 mmol) in a mixture of hexamethyldisilazane (50 ml) and chlorotrimethylsilane (5 ml) for 2h., and evaporation of the solvents], $HgBr_2$ (1.99 g; 5.5 mmol) and $CdCO_3$ (2.36 g; 16.6 mmol). The solvent is evaporated and the residue heated at 100°C for 1h. The residue is worked up as for the thymidine analogue and the product purified by column chromatography. The benzyl ether protecting groups are removed by treatment with BCl_3 as described for the 5-iodo analogue. The α,β anomers are separeted by preparative reverse phase HPLC using MeCN.H_2O as eluant.

EXAMPLE C

3'5'-Di-O-benzyl-5-bromo-2'deoxy'ß-4'-thio-L-uridine.

This compound is prepared by a method similar to the iodo compound above with the following modifications:

1. The total solvent (MeCN) volume for the reaction is 3 ml.
2. The $CdCO_3$ is omitted.
3. The excess of 5-bromo-bis-O-trimethylsilyuracil is reduced to 1.5 mole equivalents.

EXAMPLE 4

5-Bromo-2'-deoxy-4'-thio-L-uridine

The BCl_3 deprotection of the compound of Example C is conducted as for the iodo-compound.

EXAMPLE D

1-Acetoxy-3,5-di-p-toluoyl-2-deoxy-4-thio-L-erythro-pentofuranoside

A solution of benzyl 3,5-di-O-benzyl-2-deoxy-1-4-dithio-L-erythro-pentofuranoside (3.68 g; 8.76 mmol) in dry CH_2CH_2 (20ml)

is added dropwise to a stirred 1M solution of BCl_3 in CH_2Cl_2 (125 ml; 0.125 ml; 0.125 mol) at 78°C under N_2. The mixture is stirred at -78°C for 4.5h, then a mixture of MeOH-CH_2Cl_2(1:1,v/v) is added slowly. After warming to room temperature the solvents are evaporated to give the crude O-debenzylated thiosugar. The gum is dissolved in dry pyridine at 0°C under N_2 and a solution of p-toluoyl chloride (3.47 ml; 26.3 mmol) added slowly. The mixture is stirred at 0°C for 3 hours then the solvents are evaporated. The residue is dissolved in CH_2Cl_2, washed with 2M HCl, 1M Na_2CO_3 and water, dried over $MgSO_4$ and evaporated. The residue is purified by flash chromatography on SiO_2 eluted with EtOAc-hexane (1:9, v/v) to give the bis-toluoylthiosugar derivative. The product is dissolved in acetic anhydride (16 ml) and stirred at 0°C. Conc. H_2SO_4 (8 μl) is added followed after 10 min. by a second aliquot (8 μl); the reaction is monitored by TLC. After a further 55 min. stirring $NaHCO_3$ (100 mg.) is added and after 20 min. the mixture is cautiously poured into ice-water containing $NaHCO_3$. The product is extracted into CH_2Cl_2, dried and evaporated. The residue is purified by flash chromatography on SiO_2 eluted with 20-25% EtOAc-hexane.

EXAMPLE 5

2'-Deoxy-5-prop-1-ynyl-4'-thio-L-uridine

5-Prop-1-ynyluracil(0.112 g; 0.75 mmol) is heated in hexamethyl-disilazane (3 ml) containing trimethylsilyl chloride (1 ml) until the solid dissolves (4h). The solvents are evaporated and the residue dissolved in dry MeCN (6 ml). The solution is added, under N_2, to a stirred solution of the above thiosugar ester (0.2 g; 0.5 mmol) in MeCN (10 ml) at 0°C. Trimethylsilyl triflate (0.096 ml; 0.5 mmol) is added and the mixture stirred for 15 min. The mixture is diluted with CH_2Cl_2 (20 ml), poured into saturated aqueous $NaHCO_3$ and the organic layer separated. The aqueous layer is further extracted with CH_2Cl_2 and the combined organics dried and evaporated. Flash chromatography on SiO_2 eluted with EtOAc-hexane (3.2, v/v) gives the protected thionucleoside as a mixture of anomers contaminated with a little propynyluracil. This material (0.206 g; 0.397 mmol) is dissolved in MeOH (15 ml)

containing NaOMe (0.021 g; 0.397 mmol) and the mixture kept at ambient temperature overnight. The solution is neutralised with Dowex 50(H^+)ion-exchange resin, filtered and the filtrate evaporated to dryness. The solid is washed with ether (3 x 4 ml) and digested with hot acetone to give the required product. Methanol is added to the mixture and the solid filtered off to give the ß-anomer.

5-Prop-1-ynyluracil may be obtained from 5-iodouracil using the methodology analogous to that described by M.J. Robins et al (ibid).

EXAMPLE 6

2'-Deoxy-5-chloro-4'-thio-L-uridine.

Starting with 5-chlorouracil, this compound is prepared in a similar manner to that described in Example 5. 5-chlorouracil is commercially available. The compound is purified by HPLC as described above.

EXAMPLE 7

2'-Deoxy-5-trifluoromethyl-4'-thio-L-uridine:

Starting with 5-trifluoromethyluracil, this compound is prepared in a similar manner to that described in Example 5. 5-trifluoromethyluracil is commercially available.

A sample of this compound of is obtained by trituration of the crude deprotected nucleoside mixture with acetone, filtration and evaporation.

EXAMPLE 8

2'-Deoxy-5-ethynyl-4'-thio-L-uridine:

Starting with 5-ethynyluracil, this compound is prepared in a similar manner to that described in Example 5. 5-ethynyluracil may be prepared from 5-ioduracil using the methodology analogous to that described by M.J. Robins et al (ibid).

A sample of the pure ß-anomer of this compound is obtained by boiling the crude anomer mixture with MeOH and filtering off the product.

EXAMPLE 9

2'-Deoxy-5-E-(2-bromovinyl)-4'-thio-L-cytidine.

To a solution of benzyl 3,5-di-O-benzyl-2-deoxy-1,4-dithio-L-erythro-pentofuranose (4 g; 9.5 mmol) in acetic acid (50 ml) and acetic anhydride (50 ml) is added conc. sulphuric acid (50 μl) and the mixture stirred at ambient temperature for 30 min. The mixture is poured into excess sodium bicarbonate Na_2HCO_3, extracted with $CHCl_3$, the extracts dried over $MgSO_4$ and evaporated. The residue is purified by column chromatography on SiO_2 in the TLC solvent to give the 1'-acetoxy-di-O-benzylthiosugar derivative which is used directly below.

To the above derivative (0.33 g; 0.89 mmol) in dry CH_2Cl_2 (3 ml) at 0°C is added $SnCl_4$ (0.33 g; 0.89 mmol) in dry CH_2Cl_2 (3 ml) and E-5-(2-bromovinyl)-2,4-dimethoxypyrimidine (0.218 g; 0.89 mmol) in dry CH_2Cl_2 (3 ml). The stirred mixture is allowed to warm to ambient temperature and stirred for a further 4 h. The mixture is poured onto water, washed with saturated $NaHCO_3$ and dried over $MgSO_4$. After evaporation, the residue is chromatographed on SiO_2 in toluene-acetone (9:1, v/v) to give the ß-anomer of the protected thionucleoside, which is crystallised from MeOH. The above methoxy derivative of the protected thionucleoside is converted to the cytidine analogue by dissolution in NH_3/MeOH at ambient temperature for 2d. The product is isolated by column chromatography on SiO_2 eluted with $CHCl_3$-MeOH (9:1, v/v), then deprotected directly with BCl_3 as described above.

EXAMPLE 10

2'-Deoxy-5-propyl-4'thio-L-uridine:

2'-Deoxy-5-propynyl-4'thio-L-uridine, ß-anomer, (26 mg) and 5% Pd/C (40 mg) in MeOH (80 ml) is stirred in an atmosphere of hydrogen for 45 min. The mixture is filtered and evaporated to give a gum. Trituration with ether-hexane gives the product as a solid.

EXAMPLE 11

E-2'-Deoxy-5-(propen-1-yl)-4'-thio-L-uridine

(a) 5-Allyluracil.

Uracil (1 g; 9 mmol) is dissolved in water (200 ml) at 70°C and

$Hg(OAc)_2$ (2.9 g; 9.1 mmol) added. The mixture is stirred at 70°C for 1 week. After cooling to ambient temperature NaCl (1.5 g) is added and the mixture stirred for 4h. The resulting thick suspension of 5-chloromercury-uracil is filtered, the solid washed with 0.1 M NaCl solution and dried in vacuo at 85°C for 2 days. To the crude solid (1 g; 2.9 mmol) in MeCN (25 ml) is added Li_2PdCl_4 (0.76 g) and allyl chloride (2.9 ml) and the mixture stirred at ambient temperature for 1 week. The suspension is filtered and the filtrate evaporated to dryness. The residue is dissolved in MeOH (75 ml) and treated with H_2S gas; a black precipitate of HgS is removed by filtration and the filtrate is evaporated to leave a white solid. The desired product is isolated by flash chromatography on SiO_2 eluted with 8% MeOH-CH_2Cl_2 (v/v).

(b) 5-(E-propen-1-yl) uracil

To a solution of 5-allyluracil (80 mg; 0.5 mmol) in 95% aq. EtOH (50 ml) is added $(Ph_3P)_3RhCl$ (90 mg; 0.1 mmol) and the mixture heated under reflux for 3 days. The solvent is evaporated and the product isolated by flash chromatography on SiO_2 eluted with 5% MeOH-CH_2Cl_2.

(c) E-2'-Deoxy-5-(propen-1-yl)-4'-thio-L-uridine

5-(E-propen-1-yl)uracil (110 mg; 0.78 mmol) is converted to the bis-TMS-ether, coupled with the protected thiosugar and deprotected with methoxide as described for the 5-propynyl analogue. The crude product is purified by chromatography on SiO_2 eluted with 5% MeOH-CH_2Cl_2.

EXAMPLE 11

5-(2-Chloroethyl)-2'-deoxy-4'thio-L-uridine

5-(2-chloroethyl)uracil (prepared by the method J.D. Fissekis & F. Sweet, J-Org. Chem., 1973, 28, 264(0.122 g, 0.7 mmol) is added to hexamethyldisilazane (3 ml) and chloromethylsilane (0.1 ml) and the mixture is heated at reflux for 2 hours. The mixture is evaporated to dryness and to the residue is added a solution of

1-acetoxy-3,5-di-O-p-toluoyl-2-deoxy-4-thio-L-erythropentofuranoside (See Example D) (0.2 g, 0.46 mmol) in dry dichloromethane (10 ml), the mixture is cooled to 0°C with stirring and trimethylsilyltrifluoromethanesulphonate (0.1 ml) is added. After stirring at 0°C for 2 hours, dichloromethane (30 ml) is added and the reaction is quenched with a saturated solution of sodium bicarbonate (20 ml). The aqueous phase is extracted with dichloromethane (2 x 25 ml) and the combined organic phases dried (Na_2SO_4) and evaporated to dryness to give the crude product in the p-toluoyl protected form. This intermediate (0.23 g) is added to a solution of sodium methoxide (0.9 mmol) in dry methanol (20 ml) and the mixture stirred at room temperature overnight. The solution is neutralised with Dowex 50 (H^+) resin, the resin filtered and washed with methanol. The combined filtrate and washings are evaporated to dryness, the residue partitioned between ether and water, the organic layer re-extracted with water and the combined aqueous phases evaporated to dryness. The residue is chromatographed on silica gel eluting with 7% methanol/dichloromethane, the product freeze dried from water to give the title compound.

EXAMPLE 12

2'-Deoxy-5-nitro-4'-thio-L-urdine

A solution of 5-nitrouracil-2,4-bis-trimethylsilyl ether [from 5-nitrouracil (118 mg, 0.75 mmol) and hexamethyldisilazide (3 ml)-chlorotrimethylsilane (3 drops), reflux 1 h.] in dry CH_2Cl_2 (6 ml) is added to a solution of 1-acetoxy-3,5-di-p-toluoyl-2-deoxy-4-thio-L-erythro-pentofuranoside (200 mg, 0.5 mmol) in dry CH_2Cl_2 (10 ml). The stirred mixture is cooled in an ice-bath and freshly distilled trimethylsilyl trifluoromethanesulphonate (96 µl) added. After 30 min. at 0°C the mixture is poured into saturated $NaHCO_3$ (50 ml), the layers separated and the aqueous phase extracted with CH_2Cl_2. The combined CH_2Cl_2 phase is dried over $MgSO_4$, then evaporated and the residue triturated with ether to leave the crude protected nucleoside. Deprotection is accomplished by adding a solution of sodium (20 mg) in MeOH (1 ml) to a suspension of the product (140 mg) in MeOH (10 ml).

After 2 hours the solution is neutralised with Dowex 50X8 H+, filtered and evaporated. The residue is triturated with ether and the residue purified by HPLC on Zorbax C8 reversed phase eluted with MeCN-H_2O (1:9, v/v).

EXAMPLE E

Preparation of E-5-(2-bromovinyl)-Uracil-5-Bromo-2,4-dimethoxypyrimidine

A solution of 5-bromo-2,4-dichloropyrimidine (16 g: 70.2 mmol) [D.M. Mulvey et al. J. Het. Chem., 1973,p79] in dry MeOH (55 ml) was added slowly to a stirred solution of sodium (3.23 g: 140.4 mmol) in MeOH (55 ml) at 0°C over 30 min. The ice-bath was removed and the reaction mixture stirred at ambient temperature for 18 h. The precipitated salt was removed by filtration and the filtrate evaporated to give an oil. To this was added an aqueous solution of NaOH (30 ml; 30% w/v); the product separated as an upper layer and was extracted into Et_2O. The organic extracts were dried over $MgSO_4$ and evaporated. The residue was crystallised from thanol to give the product as colourless plates, yield 14.3 g, 93%, mp 62-63°C. Mass spectrum, elm/z 219 (M^+, 11%). Analysis, found: C,33.20, H,3.26, Br 36.90, N, 12.7%; $C_6H_7BrN_2O_2$ requires: C,32.90, H,3.33, Br 36.50, N, 12.80%.

5-Formyl-2,4-dimethoxypryrimidine

A solution of 1.6 M n-Buli in hexane (48 ml, 73.6 mmol) was added over 5 min. to a stirred suspension of 5-bromo-2,4-dimethoxypyrimidine (16 g; 72.9 mmol) in dry Et_2O (240 ml) at -70°C under an atmosphere of dry N_2. Dry ethyl formate (28 g: 377 mmol) was added and the orange solution stirred at -70°C for 1 h then allowed to warm slowly to ambient temperature. Water (400 ml) was added and the aqueous layer separated and extracted with Et_2O (3 x 200 ml). The ether layer was combined with the extracts and dried over $MgSO_4$, filtered and evaporated. The residue was purified by column chromatography by preloading in SiO_2 and eluting with EtOAc-hexane (3:7, v/v). Product fractions were combined and evaporated to give fine white needles, yield 6.89 g, (56%). Mass spectrum m/z 169 $(M+H)^+$ Analysis, found: C, 50:1;H,4.5;N,16.9%;$C_7H_8N_2O_3$ requires C,50.00; H,4.79; N, 16.66%

E-5-(2-carboxyvinyl)-2,4-dimethoxypyrimidine

Malonic acid (13.03 g; 126.2 mmol) and redistilled piperidine (2ml) were added to a solution of 5-formyl-2,4,dimethoxypyrimidine (10.52 g; 6.2.6 mmol) in dry pyridine (60 ml). The mixture was heated on a steam bath for 10 h then the solvent was removed by distillation under reduced pressure. The residual oil was re-evaporated from water (3 x 25 ml) and the solid thus obtained recrystallised firstly from water and then from dry methanol to give the product as white needles, yield 6.45 g; a second crop was obtained from the filtrate (1.08 g). Total yield 7.53g (57%). Mass spectrum: (El) m/z 210 (M^+). Analysis, found: C,52.1;H,4.8;N, 13.1%: $C_9H_{10}N_2O_4$ requires: C, 52.43; H, 4.79; N, 13.33%.

E-5-(2-Bromovinyl)-2,4-dimethoxypyrimidine

To a solution of E-5-(2-carboxyvinyl)-2,4-dimethoxypyrimidine (0.300 g; 1.43 mmol) in dry DMF (5 ml) was added K_2CO_3 (0.45 g: 5.25 mmol). After stirring at ambient temperature for 15 min. a solution of N.-bromosuccinimide (0.258 g; 1.45 mmol) in dry DMF (4 ml) was added dropwise over 10 min. The suspension was immediately filtered, the solid washed with DMF and the filtrate evaporated in high vacuum. The solid residue was purified by column chromatography by preloading on SiO_2 and eluting with EtOAc-hexane (7:3, v/v). Product fractions were pooled and evaporated to give fine white crystals, yield 0.561 g (45%). FAB mass spectrum: m/z245 and 247 $(M+H)^+$. Analysis, found: C,39.9; H, 3.6; N, 11.5% $C_8H_9BrN_2O_2$ requires C, 40.20; H, 3.70; N. 11.43%.

E-5-(2-Bromovinyl)uracil

To a solution of E-5-(2-bromovinyl)-2,4-dimethoxypyrimidine (2.45 g; 10 mmol) in AcOH (10 ml) was added NaI (3.3 g; 2.2 eq.; 22 mmol) and the solution heated under reflux for 3h. The hot mixture was filtered and diluted with water (15 ml). After cooling, the precipitated product was filtered off, washed with acetone (50 ml) and ether (20 ml) and dried to give a pale yellow powder (1.40 g, 65%). Mp >320°C; 60 MHz ^{1}H-NMR, DMSO-6d, δ: 7.60 (s, 1H, H-6); 7.30 (d, 1H, J=13 Hz, vinyl H); 6.80 (d, 1H, J=13 Hz, vinyl H).

EXAMPLE 13.

(A) General Experimental Procedures

Reagents (including R-(+)glycidol, *tert*-butylchlorodiphenyl silane, phenylselenyl bromide, cytosine and 5-fluorocytosine) were purchased from the Aldrich Chemical Company, Gillingham, Dorset, U.K., except 4-dimethylaminopyridine and dried tetrahydrofuran (thf) which were purchased from Fluka, Glossop, U.K. Dimethylmalonate was dried and distilled over calcium chloride, acetonitrile was similarly treated over calcium hydride; all other solvents were stored over molecular sieves when appropriate. Petrol refers to the distillate collected between 40 and 60°C. Organic solutions were dried over anhydrous magnesium sulphate.

(R)-*Tert*-butyl-diphenylsilyl glycidol

To a solution of *tert*-butylchlorodiphenyl silane (19.5 g, 70.9 mmol), imidazole (4.82 g, 70.9 mmol), and 4-dimethylamino pyridine (0.41 g, 3.3 mmol) in dichloromethane (150 ml), cooled to 0°C under nitrogen, was added the R-(+)-glycidol (5.0 g, 67.5 mmol) in a dropwise manner in 40 ml of dichloromethane. Reaction was complete after 2.5 h. The solution was washed with water, a back extraction combined, then washed with iced 1N HCl (x2), water and brine. After drying, the solution was evaporated and treated directly with the next reagent.

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 2.60 and 2.75 (ea 1H, dd, H-3), 3.10 (1H, m, H-2), 3.70 and 3.85 (ea 1H, dd, H-1), 7.40 to 7.70 (10H, m, Ar-H).

(S)-(*Tert*-butyl-diphenylsilyloxy-methyl)-thiirane

The crude (R)-silyl glycidol (67.5 mmol) was dissolved in 300 ml of methanol to which was added thiourea (5.13 g, 67.5 mmol) in one portion. The solution was stirred for 14 h, after which the solvent was evaporated. The residue was taken up in ether and washed with water (x2) and brine, then dried and evaporated. The residue was purified on a flash column eluted with neat petrol, to give the pure thiirane as a mobile oil.

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 2.10 and 2.45 (ea 1H, dd, H-3), 3.05 (1H, m, H-2), 3.55 and 3.95 (ea 1H, dd, H-1), 7.40 to 7.70 (10H, m, Ar-H).
CD Spectrum (hexane) 261 (+6.9)nm.

2-(R/S)-Carboxymethyl-4-(R)-(tert-butyl-diphenylsilyloxy-methyl)-4-thio-butanolactone

Dimethyl malonate (6.6 g. 50.1 mmol) in 75 ml of dry thf, was added dropwise to a solution of sodium bis(trimethylsilyl)amide (50.1 mmol) in 250 ml of thf at room temperature. On completion of the addition, the (S)-thiirane (13.7 g, 41.8 mmol) was added in one portion in 400 ml of thf, and the solution was refluxed for 80 h. On cooling, the solution volume was reduced and the residue partitioned between ether and saturated ammonium chloride. A second extraction was combined and washed with water then brine before drying and evaporation. The residue was columned on silica, eluted with a gradient of 10-30% ether/petrol, which gave the required carboxymethyllactone as a colourless oil.

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 2.30 and 2.70 (3H, m, H-2/3), 3.60 to 4.10 (3H, m, H-4/5), 3.75 (3H, s, OMe), 7.40 to 7.70 (10H, m, Ar-H).
Mass Spectrum (m/z) (FAB+) 429 ($M+H^+$, 25%).

4-(R)-(*Tert*-butyl-diphenylsilyloxy-methyl)-4-thio-butanolactone

The carboxymethyllactone was dissolved in 75 ml of dimethylsulphoxide, to which were added 20 drops of brine. After 2 h at 170°C the reaction was complete. After cooling, the reaction solution was directly transferred onto a pre-packed silica column, and the lactone eluted off with 30% ether in petrol, to give a colourless oil on evaporation.

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 2.05 to2.55 (4H, m, H-2/3), 3.80 (2H, m, H-5), 4.05 (1H, m, H-4), 7.40 to 7.70 (10H, m, Ar-H).

Mass Spectrum (m/z) (FAB+) 371 ($M+H^+$, 5%), 313 ($M+H-CMe_3$, 80).
CD Spectrum (EtOH) 231 (-1.2) nm.
Microanalysis, found: C, 67.73; H, 6.94; $C_{21}H_{26}O_2SSi$ requires C, 68.06; H, 7.07%.

4-(R)-(*Tert*-butyl-diphenylsilyloxy-methyl)-2-(S)-phenylselenyl-4-thio-butanolactone

The lactone (5.25 g, 14.2 mmol) in 45 ml of dry thf was added dropwise to a 1M solution of lithium bis(trimethylsilyl)amide (15.6 ml) in thf at -78°C under nitrogen (this temperature was maintained throughout the reaction). This solution was stirred for 1 h on completion of the addition, then a single portion of trimethylsilylchloride (1.69 g, 15.6 mmol) was added and stirred for 2 h. At this time, phenylselenyl bromide (3.68 g, 15.6 mmol) was added slowly in 60 ml of thf, and the reaction stirred for a further 1 h before it was allowed to warm to room temperature. The mixture was poured into water, which was extracted with three portions of ether. These combined fractions were washed twice with brine, dried and evaporated. The resulting syrup was purified by elution of a flash column with a 0-10% ether/petrol gradient, which gave the desired 2-(S)-phenylselenyl lactone, free of the small quantities of the 2-(R)-epimer which was also formed during the reaction.

^1H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 2.35 (2H, m, H-3), 3.75-3.85 (4H, m, H-2/4/5), 7.30 to 7.65 (15H, m, Ar-H).
Mass Spectrum (m/z) (FAB+) 526 ($M+H^+$, 10%).

1-O-Acetoxy-5-O-*tert*-butyl-diphenylsilyloxy-2,3-dideoxy-2-phenylselenyl-4-thio-α/β-L-ribofuranose

The lactone (7.5 mmol) was reduced by diisobutylaluminiumhydride (7.9 mmol) in 100 ml of dry toluene at -78°C over 3 h, after which the reaction was quenched with 100 ml of saturated ammonium chloride and vigorously stirred for 1 h. After filtering through a hyflo pad, the organic layer was separated and washed twice with brine, dried and evaporated. The crude lactol was dissolved

in 200 ml of dichloromethane and treated with 4-dimethylamino pyridine (1.0 g, 8.25 mmol) and acetic anhydride (0.84 g, 8.25 mmol). Reaction was complete in 2 h at room temperature, then the solution was washed sequentially with water, copper sulphate, water then brine, dried and evaporated. Purification was achieved by on a short flash column.

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.00 and 1.05 (9H, s, CMe_3), 1.95 and 2.10 (3H, 2s, CH_3CO), 2.30 to 2.70 (2H, m, H-3), 3.45 to 3.95 (4H, m, H-2/4/5), 6.10 and 6.15 (1H, 2d, H-1), 7.40 to 7.70 (15H, m, Ar-H).

(B) General procedure for glycosylation of cytosines with 1'-0-acetoxy-2'phenylselenyl-4'thio-riboside

The 1'-0-acetoxy-2'-selenyl-4'thio-riboside (2.0 mmol) was dissolved in 20 ml of acetonitrile under nitrogen. To this was added the cytosine (3.0 mmol), then potassium nonafluorobutane-1-sulphonate (6.3 mmol), hexamethyldisilazane (2.0 mmol) and trimethylsilylchloride (9.0 mmol), each in one portion. This suspension was then vigorously stirred at room temperature for 14 h, then poured into a saturated aqueous solution of sodium bicarbonate, and the mixture extracted with 3 x 50 ml portions of dichloromethane. The combined organic fractions were washed twice with brine, dried and evaporated, prior to purification on a flash silica column eluted with methanol/chloroform/ammonia (7:92.1). This purification gave the single required diastereomer as a white foam, free of small quantities of the other diastereomer formed in the reaction.

5'-0-(*Tert*-butyl-diphenylsilyl)-2,3'-dideoxy-2'-phenylselenyl-4'-thio-β-L-cytidine

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 2.15 and 2.35 (ea 1H, m, H-3'), 3.80 (4H, m, H-2'/4'/5'), 5.30 (1H, d, H-5), 6.40 (1H, d, A-H-1') 7.25 to 7.75 (1δH, m, Ar-H), 7.85 (1H, d, H-6).

5'-0-(*Tert*-butyl-diphenylsilyl)-2,3'-dideoxy-5-fluoro-2'-phenylselenyl-4'-thio-β-L-cytidine

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 2.15 and 2.35 (ea 1H, m, H-3'), 3.70 (4H, m, H-2'/4'/5'), 6.35 (1H, dd, H-1'), 7.25 to 7.75 (16H, m, Ar-H, H-6).

(C) General procedure for oxidative removal, by elimination, of phenylselenyl functionality

The 2'-selenyl nucleoside was dissolved in dry dichloromethane and cooled to -20°C under nitrogen. To this was added an equivalent of metachloroperoxybenzoic acid in one portion, and the temperature maintained during the course of the reaction (45 min). 5 eq of pyridine were then added and the solution allowed to warm to room temperature over 1 hour. After dilution with dichloromethane the solution was washed successively with water, copper sulphate (x2), sodium bicarbonate (x2), water, and brine, before drying and evaporation. Purification was achieved on a flash column, eluted with methanol/chloroform/ammonia (7:92:1).

5'-O-(*Tert*-butyl-diphenylsilyl)-2',3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 3.85 (2H, m, H-5'), 4.40 (1H, m, H-4'), 5.30 (1H, d, H-5), 5.80 and 6.20 (each 1H, m, H-2'/3'), 7.20 (1H, dd, H-1'), 7.35 to 7.70 (10H, m, Ar-H), 7.50 (1-H, d, H-6).

5'-O-(*Tert*-butyl-diphenylsilyl)-2',3'-didehydro-2',3'-dideoxy-5-Fluoro-4'-thio-β-L-cytidine

^{1}H NMR spectrum ($CDCl_3$) δ_H, 1.05 (9H, s, CMe_3), 3.80 (2H, m, H-5'), 4.40 (1H, m, H-4'), 5.75 and 6.25 (each 1H, m, H-2'/3'), 7.15 (1H, dd, H-1'), 7.35 to 7.70 (11H, m, Ar-H, H-6).

(D) General procedure for desilylation of nucleosides

The silyl ether was stirred in a thf solution with tetraethylammonium fluoride (1.1 eq) for 1 hour at room temperature when completion was evident by tlc [methanol/chloroform/ammonia (7:92:1)]. Silica gel was added to this mixture, the solvent evaporated, and the pre-absorbed

mixture placed onto a short silica column. The column was eluted with a gradient based on the tlc solvent system; the required product was either crystallised from ethanol or lyophilised.

(i) 2',3'-Didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine

^{1}H NMR spectrum (DMSO-d_6) δ_H, 3.60 (2H, m, H-5'), 4.30 (1H, m, H-4'), 5.15 (1H,brt, OH), 5.75 (1H, d, H-5), 5.85 and 6.30 (each 1H, dt, H-2'/3'), 6.85 (1H, dd, H-1'), 7.30 (2H, brd, NH_2), 7.65 (1H, d, H-6).

Infra red spectrum ν_{max} (KBr disc) 3 344, 3 199, 1 649, 1 607, 1 526, 1 499 cm^{-1}.
Mass spectrum (m/z)(FAB+)226 ($M+H^+$, 20%).
Microanalysis, found: C, 45.42, H, 4.70; N, 17.33;
$C_9H_{11}O_2N_3SF.0.14CHCl_3$ requires C, 45.41; H, 4.64, N,17.39%.
CD Spectrum (H_2O) 275 (+1.70), 231 (-6.41), 213 (+4.20)nm.

(ii) 2',3'-Didehydro-2',3'-dideoxy-5-Fluoro-4'-thio-β-L-cytidine

^{1}H NMR spectrum (DMSO-d_6) δ_H, 3.65 (2H, m, H-5'), 4.35 (1H, m, H-4'), 5.20 (1H, t, OH), 5.85 and 6.25 (each 1H, dt, H-2'/3'), 6.85 (1H, m, H-1'), 7.65 (2H, brd, NH_2), 7.95 (1H, d, H-6).
Infra red spectrum ν_{max} (KBr disc) 3 351, 3 186, 1 682, 1 647, 1 607, 1 508 cm^{-1}.
Mass spectrum (m/z)(FAB+)243 (M^+, 65%).
Microanalysis, found: C, 43.01, H, 4.26; N, 16.34;
$C_9H_{10}O_2N_3S.0.53H_2O$ requires C, 42.79; H, 4.41, N,16.63%.
CD Spectrum (EtOH) 288 (+7.62), 239 (-3.86), 214 (+5.85)nm.

EXAMPLE 14.

Methyl 2-Deoxy-3,5-di-O-(p-nitrobenzoyl)-D-threo-pentoside

To a cold (0°C) stirred solution of methyl 2-deoxy-D-erythro-pentoside (1.2g, 8.1 L) and triphenylphosphine (8.5 g, 32.4 mmol) in dry toluene (160 ml) was added a solution of p-nitrobenzoic acid in dry toluene (30 ml), followed by diisopropyl azodicarboxylate (6.4 ml, 32.4 mmol) under N_2. The reaction

mixture was allowed to warm up to room temperature and after 6 hrs of stirring the solid was filtered, the filtrate evaporated, the residue resuspended in toluene (100 ml), the precipitate filtered and filtrate evaporated to dryness the residue was purified by column chromatography eluting sequentially with ethyl-acetone hexane (3:7, 2.3 and 1:1), ethyl-acetate/toluene (3:7) and methanol/dichloro methane (1:99) to give the title compound.

NHR: ('H) δ ($CDCl_3$): 8.4-8.1 (8H, m, aromatic), 5.85 (1H, m, H-3), 5.3 (0.64H, dd, H-1, α-anomer), 5.2 (0.36H, dd, H-1, β anomer), 4.75-4.55 (3H, m, H-4, H-5), 3.48 (3H, s, OMe), 3.43 (3H, s, OMe), 2.65-2.25 (2H, m, H-2).

MASS SPECTRUM

FAB 447 (H+1); NBA Matrix

2-Deoxy-3,5-di-O-(p-nitrobenzoyl)-D-threopentose di-(p-methoxybenzyl) dithioacetal

To a stirred solution of methyl 2-deoxy-3,5-di-O-(p-nitrobenzyol)-D-threopentoside (218 mg, 0.488 mmol) in dry toluene (40 ml) was added p-methoxybenzylmercaptan (218 μl; 2.44 mmol) followed by titanium tetrachloride (54 μl, 0.488 mmol) at room temperature under N_2. After 20 mins. the reaction mixture was quenched with $NaHCO_3$ (100 ml) and extracted with ether (3 x 40 ml). The combined organic layers were dried (Na_2SO_4), evaporated to dryness and the residue was purified by column chromatography eluting with ethyl-acetate/hexane (1:4-2:3) to give the title compound.

NMR Spectrum

('H) δ ($CDCl_3$) : 8.32-7.9 (8H, m, aromatic), 7.2-7 (4H, dd, aromatic), 6.8-6.55 (4H, dd, aromatic), 5.52 (1H, m, H-3), 4.35 (2H, m, H-5), 3.95 (1H, m, H-4), 3.85-3.6 (10H, m, $PhCH_2$ S,OMe), 3.52 (1H, m, H-1), 2.5-2.1 (2H, m, H-2).

MASS SPECTRUM

FAB - 723 (M+1) NBA - Matrix

2-Deoxy-3,5-di-O-(p-nitrobenzoyl)-4-O-Methanesulphonyl-D-threo-Pentose-Di-(p-methoxybenzyl) dithioacetal

To a cold (0°C) stirred solution of 2-deoxy-3,5-di-O-(p-nitrobenzoyl)-D-threopentose-di-(p-methoxybenzyl) dithioacetal (1.45g, 2 mmol) in dry pyridine (20 ml) was added methanesulphonyl chloride (188 μl, 2.44 mmol) under N_2. After stirring at room temperature for 5 hrs. the reaction mixture was quenched with water, the solvent evaporated to dryness under high vacuum and the residue was partitioned between dichloromethane/water (100 ml / 75 ml). The aqueous layer was further extracted with dichloromethane (3 x 50 ml), the combined organic layers dried (Na_2SO_4) and evaporated to dryness. Residual pyridine was co-evaporated with ethanol (2 x 50 ml) to give the title compound.

NMR SPECTRUM

H) δ ($CDCl_3$): 8.4-7.9 (8H, m, aromatic), 7.3-7 (4H, dd, aromatic), 6.8-6.5 (4H, dd, aromatic), 5.3 (1H, m, H-3), 5.0 (1H, m, H-4), 4.65-4.5 (1H, m, H-5), 4.45-4.3 (1H, m, H-5), 3.9-3.6 (10H, m, PH$\underline{CH_2}$ S, O$\underline{Me}$), 3.55 (1H, dd, H-1), 3.0 (3H, s, O$\underline{Ms}$), 2.5-2 (2H, m, H-2).

p-Methoxybenzyl-2-deoxy-3,5-di-O-(p-Nitrobenzoyl)-1,4-dithio-L-erythro-pentofuranose

To a stirred solution of 2-deoxy-3,5-di-O-(p-nitrobenzoyl)-4-O-methanesulphonyl-D-threo-pentose-di-(p-methoxybenzyl)-dithioacetal (1.46 g, 1.82 mmol) in dry dimethylformamide (40 ml) was added triethylamine (381 μl, 2.73 mmol) followed by sodium iodide (2.2 g, 14.6 mmol) at room temperature under N_2. After 20 hrs. of stirring at 100°C the reaction mixture was quenched with water, the solvent evaporated to dryness under high vacuum and the residue was partitioned between dichloromethane and water. The organic layer was washed with water (2 x 30 ml) dried

(Na_2SO_4), evaporated to dryness and the residue was purified by column chromatography eluting with ethyl-acetate/hexane (3:7) to give the title compound (5).

NMR SPECTRUM

(H) δ ($CDCl_3$): 8.3-8.1 (8H, m, aromatic), 7.35-7.2 (2H, d, aromatic), 6.9-6.8 (2H, d, aromatic), 5.8 (1H, m, H-3), 4.65-4.4 (3H, m, H-1, H-5), 4-3.75 (6H, m, H-4, Ph$\underline{CH_2}$S, O$\underline{Me}$ 2.7-2.3 (2H, m, H-2).

2'-Deoxy-3',5'-di-O-(p-nitrobenzoyl)-5-fluoro-4'-thio-L-1,B-cytidine

A mixture of 5-fluorocytosine (42 mg, 0.324 mmol) and bis (trimethylilyl)-acetamide (133 μl, 0.43 mmol) in dry acetonitrile (15 ml) was stirred at 80°C under N_2. After 1 hr. the solution was cooled to room temperature and to it was added dropwise a solution of p-methoxybenzyl-2-deoxy-3,5-di-O-(p-nitrobenzoyl)-1,4-dithio-L-erythro-pentofuranose (158 mg, 0.27 mmol) in dry acetonitrile (10 ml), followed by N-iodosuccinimide (61 mg 0.27 mmol) in dry acetonitrile (5 ml) and trimethylsilyltrifluoromethane sulphonate (52 μl, 0.27 mmol). After stirring for 2 hrs. at room temperature TLC (ethyl-acetate/hexane 3.7) showed presence of starting material. A further amount of N-iodosuccinimide (6 mg, 0.027 mmol) was added and after stirring overnight the reaction mixture was diluted with dichloromethane (30 ml), quenched with 10% $NaHCO_3$ (30 ml) and to the mixture was added a solution of sodiumthiosulphate (30m m). The organic layer was separated and the aqueous layer was further extracted with dichloromethane (2 x 30 ml) and the combined organic layers were washed with water (3 x 30 ml), dried (Na_2SO_4) and evaporated to dryness. The resulting residue was purified by column chromatography eluting with methanol/dichloromethane (1:9) to give title compound (6) as an anomeric mixture $\alpha:\beta$ = 1.8:1.

NMR SPECTRUM

('H) δ (d_6DMSO):8.45-8 (9H, m, aromatic, H-6), 7.9-7.5 (2H, m, NH_2), 6.45 (0.35H,t, H-1', β anomer), 6.25 (0.65H, m, H-1', α anomer), 5.9-5.65 (1H, m, H-3'), 4.8 -4.5 (2H m, H-5), 3.65 (1H, m, H-4'), 3-2.5 (2H, m, H-2').

2'-deoxy-5-fluoro-4'-thio-L-α,β-cytidine

To a stirred solution of 2'-deoxy-3',5'-di-O-(p-nitrobenzoyl)-5-fluoro-4'-thio-L-α,B-cytidine (55.6 mg, 0.01 mmol) was added a 30% w/v solution of sodium methoxide in methanol (360 μl 0.198 mmol) at room temperature under N_2. After 3 hrs. TLC in methanol/dichlromethane (3:17) showed presence of product and starting material. The reaction mixture was neutralised with "DOWEX" 50W-X8 (H) to pH 7, filtered washed with methanol and evaporated to dryness. The residue was purified by column chromatography, eluting with methanol/dichloromethane (3:17 and 1:4) to give the first batch of product. The recovered starting material was deprotected with 30% w/v solution of sodium methoxide in methanol (180μl; 0.09 mmol) and worked up as above. The combined products were purified by HPLC (ZORBAX C8, using a O-35% 20 minute gradient of 10% H_2O/ACN in 0.1M NH_4OAc pH4.0, to give the title compound (7) as an anomeric mixture α:β=1.2.

NMR - SPECTRUM

('4) δ (d_4-MeOH) 8.5 (0.35H, d, H-6, α-anomer), 8.4 (0.65H, d, H-6, β-anomer), 6.35 (0.65H, m, H-1', β anomer), 6.25 (0.35H, m, H-1', α-anomer), 4.6-4.35 (1H, m, H-3'), 3.8-3.4 (3H, m, H-4', H-5'), 2.6-2.0 (2H, m, H-2').

MASS SPECTRUM

E170 MI=261

BIOLOGICAL DATA

a) Anti-HSV Activity

Herpes Simplex Virus types 1 (HSV 1) and 2 (HSV2) were assayed in monolayers of Vero cells in multiwell trays. The virus strains used were SC16 and 186 for HSV-1 and HSV-2 respectively. Activity of compounds was determined in the plaque reduction assay, in which a cell monolayer was infected with a suspension of the appropirate HSV, and then overlaid with nutrient agarose in the form of a gel to ensure that there was no spread of virus throughout the culture. A range of concentrations of compound of known molarity was incorporated in the nutrient agarose overlay. Plaque numbers at each concentration were expressed as percentages of the control and a dose-response curve was drawn.

b) Anti-CMV Activity

Human cytomogalovirus (HCMV) was assayed in monolayers of either MRC5 cells (human embryonic lung) in multiwell trays. The standard CMV strain AD 169 was used. Activity of compounds is determined in the plaque reduction assay, in which a cell monolayer is infected with a suspension of HCMV, and then overlaid with nutrient agarose in the form of a gel to ensure that there is no spread of virus throughout the culture. A range of concentrations of compound of known molarity was incorporated in the nutrient agarose overlay. Plaque numbers at each concentration of drug are expressed as percentage of the control and a dose-response curve is drawn.

c)Anti-VZV Activity

Clinical isolates of varicella zoster virus (VZV) were assayed in monolayers of MRC-5 cells. MRC-5 cells are derived from human embyonic lung tissue. A plaque reduction assay was used in which a suspension of the virus stock was used to infect monolayers of the cells in multiwell trays. a range of concentrations of the compound under test of known molarity was added to the wells. Plaque numbers at each concnetration were expressed as percentages of the control and a dose response curve was constructed. From these curves the 50% inhibitory

concentration of each drug was determined.

d) HBV Assay (Method 1)

Materials Virus/Cells

The cell line used was derived from a hepatoblastoma cell line, Hep G2, which had been transfected with a plasmid containing four 5'-3' tandem copies of the hepatitis B virus genome, subtype ayw, to produce the cell line designated 2:2:15. (Sells *et al* PNAS 84 1005-1009, 1987). These cells carry the Hep B DNA both as chromosomally integrated sequences and episomally. The cells constitutively produce small amounts of virus particles. A higher virus producing clone P5A, was obtained from the 2.2.15 cells for use in the assay.

Media

Cells were grown in RPM1 1640 containing 0.5% penicillin and streptomycin, 2mML-glutamine and 10% foetal calf serum.

Methods

Assays were performed in 24 well plates which were seeded with approximately $2.5x10^4$ cells/well and grown for 5 days at 37°C in 5% CO_2, the monolayers were then incubated with RPM1 1640, 0.5% penicillin and streptomycin, 2mM L-glutamine and 2% FCS containing the test compounds at the required concentrations. Medium was replaced every 48 hours with fresh medium containing the test compound. The plates were incubated for 10 days, the medium was removed and the cells scraped from the wells in 0.5ml of PBS, the cells were pelleted at 5000 rpm for 5 minutes the supernatant discarded and the cells frozen at -20°C. The cells were thawed and resuspended in 500µl of lysis buffer (150 mM NaCl, 20mM Tris/HCl pH7.4, 10mM EDTA and 0.6% SDS) and 50 µL of proteinase K (20mg/ml) added and the samples incubated at 37°C for 2 hours. DNA was extracted on an Autogen 540 DNA extractor and dissolved in a final volume of 50µl of water. DNA was digested with the restriction enzyme Hind III at 37°C for 16 hours and the DNA fragments separated on 1% agarose gel. The separated DNA was transferred by capillary blotting to hybond N^+ nylon membrane (Amersham International) and, after prehybridisation, hybridized with a ^{32}P labelled positive strand RNA transcript of the core region of the hepatitis B genome, subtype ayw, at 42°C overnight in the presence of 50% formamide. After extensive washing the

blot was exposed to X-ray film and the intensity of the hybridization to the replicative intermediate DNA analysed by a Milli Pore 610 imager. Results were compared to a control sample containing no test compound.

e) HBV Assay (Method 2)

Anti-HBV activity of compounds of formula (I) was determined with a high capacity assay for assessing efficacy. Supernatants from growing HBV-producing cells (HepG2 2.2.15, P5A cell line) in 96-well plates are applied to microtiter plate wells which have been coated with a specific monoclonal antibody to HBV surface antigen (HBsAg). Virus particles present in the supernatants bind to the antibody and remain immobilized while other debris is removed by washing. These virus particles are then denatured to release HBV DNA strands which are subsequently amplified by the polymerase chain reaction and detected with a colorimetric hybrid-capture assay. Quantitation is achieved through fitting of a standard curve to dilutions of a cell supernatant with known HBV DNA content. By comparing HBV DNA levels of untreated control cell supernatants with supernatants containing a compound of formula (I), a measure of anti-HBV effectiveness is obtained.

Immunoaffinity Capture of HBV

HBV producer cells, 2500 cells/well, were seeded in 96-well culture dishes in RPMI/10% fetal bovine serum/2mM glutamine (RPMI/10/2). Media were replenished on days 1, 3, 5 and 7 with dilutions of a compound of formula (I) in RPMI/10/2 to a final volume of 150 µl. Fifty µL of mouse monoclonal anti-HBsAG antibody (10µg/mL in PBS) were added to each well of a round-bottom microtiter plate. After incubation overnight at 4°C, the solutions were aspirated and replaced with 100 µL of 0.1% BSA in PBS. Samples were incubated for 2 hours at 37°C and washed three times with PBS/0.01% Tween-20 (PBS/T) using a Nunc Washer. Ten µL of 0.035% Tween 20 in PBS were then added to all wells by Pro/Pette. Cell supernatants (25 µL) containing extracellular virion DNA were transferred into wells by Pro-Pette; the final Tween concentration is 0.01%. Twenty-five µL HBV standard media

dilutions in RPMI/10/2 were added to 2 rows of wells to serve as an internal standard curve for quantitation, and the plates were sealed and incubated at 4°C overnight. Samples were washed 5 times with PBS/T and 2 times with PBS, aspirating the last wash. Next, 25 µL of 0.09N NaOH/0.01% NP40 were added to each well by Pro/Pette, and the sample wells were sealed and incubated at 37°C for 60 minutes. Samples were then neutralized with 25 µL of 0.09N HCI/100 mM tris (pH 8.3).

Polymerase Chain reaction (PCR)

Polymerase chain reaction (Saiki, R.K. *et al*, Science, 239 (4839) 487-91 (1988)) was carried out on 5µL samples, using a Perkin Elmer PCR kit. PCR is performed in "MicroAmp tubes" in a final volume of 25 µL. Primers were chosen from conserved regions in the HBV genome, as determined by alignment of several sequences. One primer is biotinylated at the 5-prime end to facilitate hybrid-capture detection of the PCR products. All primers were purchased from Synthecell Corp, Rockville, MD 20850.

Hybrid-Capture Detection of PCR Products

PCR products were detected with horse radish peroxidase-labelled oligonucleotide probes (Synthecell Corp, Rockville, MD 20850), which hybridize to biotinylated strands of denatured PCR products directly in streptavidin-coated microtiter plate wells, using essentially the method of Holodiniy, M *et al*, Bio Techniques, 12 (1) 37-39 (1992). Modifications included the use of 25k PCR reaction volumes and sodium hydroxide denaturation instead of heat. Simultaneous binding of the biotin moiety to the plate-bound streptavidin during the hybridization serves to "capture" the hybrids. Unbound labelled probes were washed away before colorimetric determination of the bound (hybridized) horse radish peroxidase. Quantities of HBV DNA present in the original samples were calculated by comparison with standards. These values were then compared to those from untreated cell cultures to determine the extent of anti-HBV activity.

IC_{50} (the median inhibitory concentration) is the amount of compound which produces a 50 percent decrease in HBV DNA.

f) HeLa-CD4$^+$ cell assay for evaluating susceptibility of HIV to antiviral compounds

Susceptibility of HIV to inhibitors was determined by infection of HT4-6C cell monolayers as described by Larder, B.A. Chesebro, B & Richman, D.D. Antimicrob. Agents Chemother. 1990 34, 436-441. Briefly cells were seeded in 24-well multiwells at 5 x 10^4 cells per well and incubated overnight at 37°C in growth medium (DMEM10). Monolayers were infected with 100-200pfu of cell-free virus in 0.2ml of DMEM containing 5% fetal bovine serum plus antibiotics (DMEM5) and incubated for 1 hour at 37°C to allow virus adsorption. Following this time 0.3m, of DMEM5 (with or without inhibitor) was added to each well and cultures were incubated at 37°C for 2-3 days. Monolayers were fixed with 10% formaldehyde solution in PBS and stained with 0.25% crystal violet in order to visualize virus plaques. Individual foci of multinucleated gian cells (plaques) were apparent using this staining procedure. ID_{50} values were derived from plots of percent plaque reduction versus inhibitor concentration.

g) MT4/MTT dye uptake assay

100μl of RPMI growth medium, with or without inhibitor, was added to each well of a 96-well microplate. MT4 cells, either mock-infected or infected with HIV at a m.o.i. of 0.01 for 1 hour at 37°C then washed three times, were added to the plate at a concentration of 40,000 cells per well, and the cultures incubated at 37°C for 5 days. 20μl of a 5mg/ml solution of 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-tetrazolium bromide (MTT) was added to each well and the panels incubated at 37°C for 2 hours. Acidified isopropanol (AIP) was then added (170μl/well) and the plates maintained at room temperature overnight prior to reading spectrophotometrically at 590 nm. Viable cells are able to reduce the yellow MTT to its purple formozan product, which is solubilized by the AIP, while wells containing killed cells remain yellow. The concentration of drug (IC_{50}) required to protect 50% of the cells from viral killing was determined from regression analysis of percentage cell death against drug concentration.

h) Cell Toxicity

Cell toxicity is assessed in cell growth inhibition assay. Subconfluent cultures of Vero cells grown on 96-well microtiter dishes are exposed to different dilutions of drug, and cell viability determined daily on replicate cultures using uptake of a tetrazolium day (MTT). The concentration required for 50% inhibition of cell viability at 96 hours is termed $CCID_{50}$.

Biological test results.

The compounds 2',3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine and 2',3'-didehydro-2',3'-dideoxy-5-fluoro-4'-thio-β-L-cytidine were tested for activity against HBV using both assay methods described above, and against HIV using the assay described above. The results of the tests, together with toxicity data (derived as described in (g) above are shown in Table 1.

TABLE 1

COMPOUND	HBV Hep G2 cells (uM)			HIV (uM)		CCID50
	METHOD 1 IC_{50}	METHOD 2 IC_{50}	TOX	HeLa CD4	MT4	(Vero)
2',3'-didehydro-2'3'-dideoxy-4'-thio-β-L-cytidine	<4	3.4, 3.6	>200	0.4	6	>500
2'3'-didehydro-2'3'-dideoxy-5-fluoro-4'-thio-β-L-cytidine	1.2	0.32 (60%); 0.88	>200	0.85	3.6	>500

EXAMPLES

The following examples illustrate pharmaceutical formulations according to the invention in which the active ingredient is a compound of formula (I).

Formulation Example A Tablet

Active ingredient	100 mg
Lactose	200 mg
Starch	50 mg
Polyvinylpyrrolidone	5 mg
Magnesium stearate	4 mg
	359 mg

Tablets are prepared from the foregoing ingredients by wet granulation followed by compression.

Formulation Example B Opthalmic Solution

Active ingredient	0.5 g
Sodium chloride, analytical grade	0.9 g
Thiomersal	0.001 g
Purified water to:	100 ml
pH adjusted to:	7.5

Formulation Example C: Tablet Formulations

The following formulations a and b are prepared by wet granulation of the ingredients with a solution of povidone, followed by addition of magnesium stearate and compression.

Tablet Formulation a

	mg/tablet	mg/tablet
(a) Active ingredient	250	250
(b) Lactose B.P.	210	26
(c) Povidone B.P	15	9
(d) Sodium Starch Glycolate	20	12
(e) Magnesium Stearate	5	3
	500	300

Tablet Formulation b

	mg/tablet	mg/tablet
(a) Active ingredient	250	250
(b) Lactose	150	-
(c) Avicel PH 101	60	26
(d) Povidone B.P.	15	9
(e) Sodium Starch Glycollate	20	12
(f) Magnesium Stearate	5	3
	500	300

Tablet Formulation c

	mg/tablet
Active ingredient	100
Lactose	200
Starch	50
Povidone	5
Magnesium stearate	4
	359

The following formulations, D and E, are prepared by direct compression of the admixed ingredients. The lactose used in formulation E is of the direct compression type.

Tablet Formulation d

	mg/capsule
Active Ingredient	250
Pregelatinised Starch NF15	150
	400

Tablet Formulation e

	mg/capsule
Active Ingredient	250
Lactose	150
Avicel	100
	500

Tablet Formulation f (Controlled Release Formulation)

The formulation is prepared by wet granulation of the ingredients

(below) with a solution of povidone followed by the addition of magnesium stearate and compression.

	mg/tablet
(a) Active Ingredient	500
(b) Hydroxpropylmethylcellulose (Methocel K4M Premium)	112
(c) Lactose B.P.	53
(d) Povidone B.P.C.	28
(e) Magnesium Stearate	7
	700

Drug release takes place over a period of about 6-8 hours and was complete after 12 hours.

Formulation Example D: Capsule Formulations

Capsule Formulation a

A capsule formulation is prepared by admixing the ingredients of Formulation D in Example C above and filling into a two-part hard gelatin capsule. Formulation B (infra) is prepared in a similar manner.

Capsule Formulation b

	mg/capsule
(a) Active ingredient	250
(b) Lactose B.P.	143
(c) Sodium Starch Glycollate	25
(d) Magnesium Stearate	2
	420

Capsule Formulation c

	mg/capsule
(a) Active ingredient	250
(b) Macrogol 4000 BP	350
	600

Capsules are prepared by melting the Macrogol 4000 BP, dispersing the active ingredient in the melt and filling the melt into a two-part hard gelatin capsule.

Capsule Formulation d

	mg/capsule
Active ingredient	250
Lecithin	100
Arachis Oil	100
	450

Capsules are prepared by dispersing the active ingredient in the lecithin and arachis oil and filling the dispersion into soft, elastic gelatin capsules.

Capsule Formulation e (Controlled Release Capsule)

The following controlled release capsule formulation is prepared by extruding ingredients a, b, and c using an extruder, followed by spheronisation of the extrudate and drying. The dried pellets are then coated with release- controlling membrane (d) and filled into a two-piece, hard gelatin capsule.

	mg/capsule
(a) Active Ingredient	250
(b) Microcrystalline Cellulose	125
(c) Lactose BP	125
(d) Ethyl Cellulose	13
	513

Formulation Example E: Injectable Formulation

Active ingredient	0.200 g
Sterile, pyrogen free phosphate buffer, pH 7.0 to	10 ml

The active ingredient is dissolved in most of the phosphate buffer (35-40°C), then made up to volume and filtered through a sterile micropore filter into a sterile 10ml amber glass vial (type 1) and sealed with sterile closures and overseals.

Formulation Example F:	**Intramuscular injection**
Active Ingredient	0.20 g
Benzyl Alcohol	0.10 g
Glucofurol 75	1.45 g
Water for Injection q.s. to	3.00 ml

The active ingredient is dissolved in the glycofurol. The benzyl alcohol is then added and dissolved, and water added to 3 ml. The mixture is then filtered through a sterile micropore filter and sealed in sterile 3 ml glass vials (type 1).

Formulation Example G:	**Syrup Suspension**
Active ingredient	0.2500 g
Sorbitol Solution	1.5000 g
Glycerol	2.0000 g
Dispersible Cellulose	0.0750 g
Sodium Benzoate	0.0050 g
Flavour, Peach 17.42.3169	0.0125 ml
Purified Water q.s. to	5.0000 ml

The sodium benzoate is dissolved in a portion of the purified water and the sorbitol solution added. The active ingredient is added and dispersed. In the glycerol is dispersed the thickener (dispersible cellulose). The two dispersions are mixed and made up to the required volume with the purified water. Further thickening is achieved as required by extra shearing of the suspension.

Formulation Example H:	**Suppository**
	mg/suppository
Active Ingredient (63μm)*	250
Hard Fat, BP (Witepsol H15 - Dynamit NoBel)	1770
	2020

* The active ingredient is used as a powder wherein at least 90% of the particles are of 63μm diameter or less.

One-fifth of the Witepsol H15 is melted in a steam-jacketed pan at 45°C maximum. The active ingredient is sifted

through a 200μm sieve and added to the molten base with mixing, using a silverson fitted with a cutting head, until a smooth dispersion is achieved. Maintaining the mixture at 45°C, the remaining Witepsol H15 is added to the suspension and stirred to ensure a homogenous mix. The entire suspension is passed through a 250μm stainless steel screen and, with continuous stirring, is allowed to cool to 40°C. At a temperature of 38°C to 40°C 2.02g of the mixture is filled into suitable plastic moulds. The suppositories are allowed to cool to room temperature.

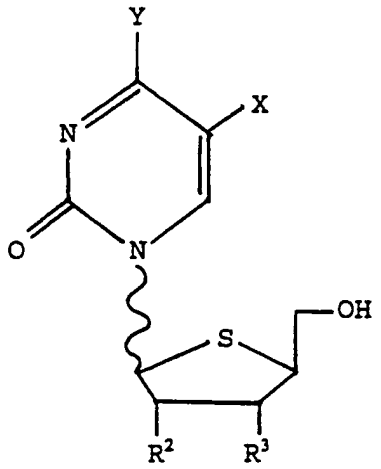
Formulation Example I: Pessaries

	mg/pessary
Active ingredient 63μm	250
Anydrate Dextrose	380
Potato Starch	363
Magnesium Stearate	7
	1000

The above ingredients are mixed directly and pessaries prepared by direct compression of the resulting mixture.

CLAIMS

1. A pyrimidine 4'-thio-L-nucleoside of the general formula (I):



wherein: Y is hydroxy or amino;
X is hydrogen, hydroxy, mercapto, halo, trifluoromethyl, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, hydroxyC_{1-3}alkyl, formyl, C_{2-6}alkenyl, C_{2-6} haloalkenyl, C_{2-6} alkynyl, C_{1-6}alkoxy, C_{1-6}alkylthio, C_{1-6}alkoxyC_{1-2}alkyl, C_{1-6}alkylthiomethyl, amino, monoC_{1-6}alkylamino, diC_{1-6}alkylamino, cyano, thiocyanate or nitro; R^2 is hydrogen and R^3 is hydroxy or hydrogen, or together R^2 and R^3 form a carbon-carbon bond;
and physiologically functional derivatives thereof.

2. A compound according to claim 1 in which X is hydrogen, hydroxy, mercapto, halo, trifluoromethyl, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, hydroxyC_{1-3}alkyl, formyl, C_{2-6} alkenyl, C_{2-6} haloalkenyl, C_{2-6} alkynyl, C_{1-6}alkoxy, C_{1-6}alkoxyC_{1-2}alkyl, amino, monoC_{1-6}alkylamino, diC_{1-6}alkylamino, cyano or nitro.

3. A compound according to claim 1 or 2 in which X is hydrogen, halo, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, C_{2-6} alkenyl, C_{2-6} haloalkenyl, C_{2-6} alkynyl, cyano or nitro.

4. A compound according to any one of claims 1 to 3 wherein X is fluoro, C_{2-3} alkyl, C_{3-4} alkenyl, halovinyl or C_{3-4} alkynyl.

5. A compound according to any one of claims 1 to 4 wherein Y is amino.

6. A compound according to any one of claims 1 to 5 wherein R^2 and R^3 form a carbon-carbon bond.

7. A compound according to claim 1 wherein the pyrimidine 4'-thio-L-nucleoside is:
2'-deoxy-4'-thio-L-uridine,
2'-deoxy-4'-thio-L-cytidine,
2'-deoxy-5-fluoro-4'-thio-L-cytidine
2'-deoxy-5-methyl-4'-thio-L-uridine,
5-(2-chloroethyl)-2'-deoxy-4'-thiouridine;
5-nitro-2'-deoxy-4'-thiouridine;
5-amino-2'-deoxy-4'-thiouridine;
5-methylamino-2'-deoxy-4'-thiouridine;
E-5-(2-bromovinyl)-2'-deoxy-4'-thio-L-uridine,
2'-deoxy-5-iodo-4'-thio-L-uridine,
5-bromo-2'-deoxy-4'-thio-L-uridine,
5-chloro-2'-deoxy-4'-thio-L-uridine,
2'-deoxy-5-ethyl-4'-thio-L-uridine,
2'-deoxy-5-prop-1-ynyl-4'-thio-L-uridine,
2'-deoxy-5-fluoro-4'-thio-L-uridine,
2'-deoxy-5-trifluoromethyl-4'-thio-L-uridine,
2'-deoxy-5-ethynyl-4'-thio-L-uridine,
2'-deoxy-5-E-(2-bromovinyl)-4'-thio-L-cytidine,
2'-deoxy-5-propyl-4'-thio-L-uridine,
E-2'-deoxy-5-(propen-1-yl)-4'-thio-L-uridine,
1-(2,3-didehydro-2,3-dideoxy-4-thio-L-ribofuranosyl)-5-methyluracil,
1-(2,3-dideoxy-4-thio-L-ribofuranosyl)-5-methyluracil,
2',3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine,
2',3'-didehydro-2',3'-dideoxy-5-fluoro-4'-thio-β-L-cytidine,
5-bromo-2'3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine,
5-chloro-2',3'-didehydro-2',3'-dideoxy-4'-thio-β-L-cytidine, or
2',3'-didehydro-2',3'-dideoxy-5-iodo-4'-thio-β-L-cytidine.

8. A compound according to any one of claims 1 to 7 wherein the pyrimidine 4'-thio-L-nucleoside is the ß-anomer.

9. A physiologically functional derivative of a pyrimidine nucleoside of Formula (I) according to any one of

claims 1 to 8.

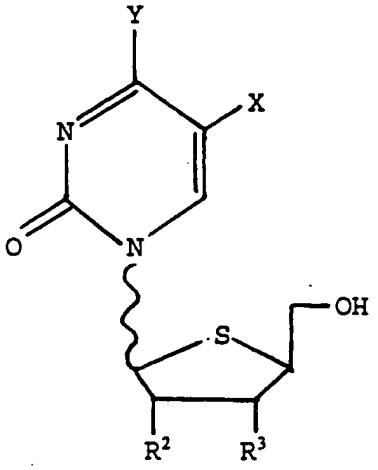
10. A derivative according to claim 9 which is an alkali metal, alkali earth metal, ammonium, tetra (C_{1-4} alkyl) ammonium, hydrochloride or acetate salt, or a mono- or di-carboxylic acid ester or an alkali metal, alkali earth metal, ammonium or tetra (C_{1-4}) alkyl ammonium salt.

11. A composition comprising a compound according to any one of claims 1 to 10 in association with a pharmaceutically acceptable carrier or diluent.

12. A compound according to any one of claims 1 to 10 or a composition according to claim 11 for use in a method of treatment or prophylaxis of virus infections.

13. Use of a compound according to any one of claims 1 to 10 or a composition according to claim 11 for the manufacture of a medicament for use in the treatment or prophylaxis of virus infections.

14. A process for the production of a pyrimidine 4'-thio-L-nucleoside of the formula (I):



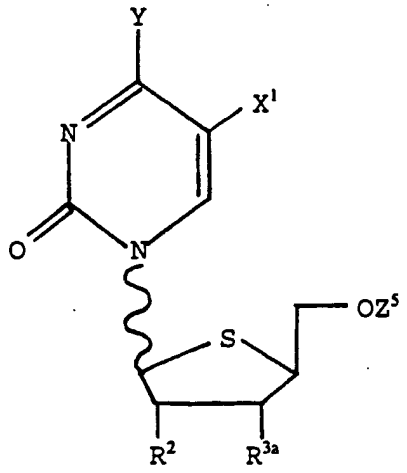
wherein Y is hydroxy or amino;

X is hydrogen, hydroxy, mercapto, halo, trifluoromethyl, methyl, C_{2-6}alkyl, C_{1-6}haloalkyl, hydroxyC_{1-3}alkyl, formyl, C_{2-6} alkenyl, C_{2-6} haloalkenyl, C_{2-6} alkynyl, C_{1-6}alkoxy, C_{1-6}alkylthio, C_{1-6}alkoxyC_{1-2}alkyl, C_{1-6}alkylthiomethyl, amino, monoC_{1-6}alkylamino, diC_{1-6}alkylamino, cyano, thiocyanate or nitro; R^2 is hydrogen and R^3 is hydroxy or hydrogen, or together R^2 and R^3 form a carbon-carbon bond;

and physiologically functional derivatives thereof, which process

comprises:

A) reacting a compound of formula (II)



(II)

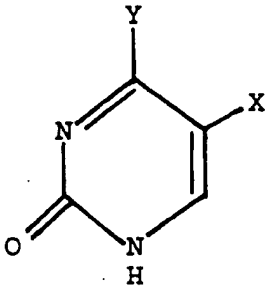
wherein X^1 is a precursor for the group X as defined in relation to formula (I);

Y and R^2 are as defined in relation to formula (I);

R^{3a} either forms a carbon-carbon double bond with R^2 or when R^2 is H, R^{3a} is hydrogen, hydroxy or a group OZ^3 where Z^3 is a hydroxyl protecting group; and

Z^5 is hydrogen or a hydroxyl-protecting group, with a reagent or reagents serving to convert the group X^1 to the desired group X; or

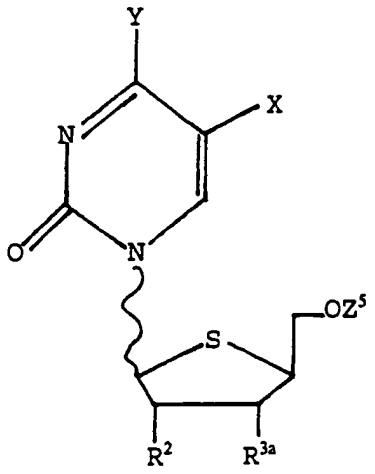
B) reacting a compound of formula (III)



(III)

wherein X and Y are as defined in relation to formula (I) or a protected form thereof with a 4-thio sugar compound serving to introduce the 4-thio sugar moiety, or a protected form thereof, at the 1-position of the compound of formula (III); or

C) reacting a compound of formula (IV)



(IV)

wherein X and Y are as defined in relation to formula (II), Z^5 is a hydroxy protecting group or hydrogen; R^2 and R^{3a} are as defined above wherein at least one of R^{3a} and Z^5 represents a precursor group for the group(s) R^3 and/or R^5 in formula (I) under conditions or with a reagent serving to convert the groups R^{3a} and/or Z^5 into the respective groups R^3 and/or H;

and, where necessary or desired, thereafter optionally effecting one or more of the following further steps in any desired or necessary order:

a) removing each of the protecting groups,

b) converting a compound of formula (I) or a protected form thereof into a further compound of formula (I) or a protected form thereof,

c) converting the compound of formula (I) or a protected form thereof into a physiologically acceptable derivative of the compound of formula (I) or a protected form thereof,

d) converting a physiologically acceptable derivative of the compound of formula (I) or a protected form thereof into the compound of formula (I) or a protected form, thereof,

e) converting a physiologically acceptable derivative of the compound of formula (I) or a protected form thereof into another physiologically acceptable derivative of the compound of formula (I) or a protected form thereof,

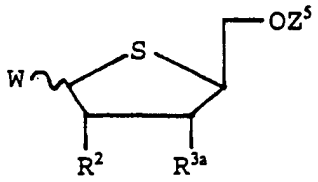
f) performing an anomerisation reaction in order to convert an

α-anomer of a compound of formula (I) into a β-amoner or to convert a β-anomer of a compound of formula (I) into an α-anomer, and

g) where necessary, separating the α and ß anomers of the compound of formula I or a protected derivative thereof or of a physiologically acceptable derivative of a compound of formula (I) or a derivative thereof.

15. A process according to claim 14 for the production of a compound according to any one of claims 2 to 10.

16. A process according to claim 14 or claim 15 wherein the 4-thio sugar derivative is a compound of formula (V)

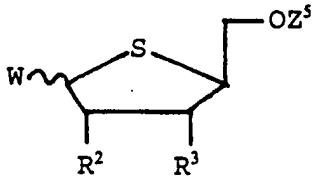


(V)

wherein R^2, R^{3a} and Z^5 are as defined in claim 14 and W is a leaving group.

17. A process according to any one of claims 14 to 16 wherein the 4-thio sugar derivative is a 1-acetoxy-4-thio sugar derivative.

18. A compound of the formula (V)



(V)

wherein R^2 is hydrogen, R^3 is a group OZ^3 where Z^3 is a hydroxyl protecting group, Z^5 is a hydroxyl protecting group and and W is -S-CH_2-Ar where Ar is an optionally substituted aryl group.

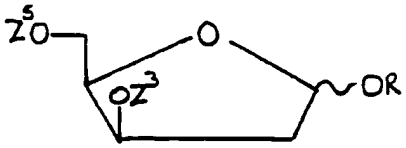
19. A compound according to claim 18 wherein Z^3 and Z^5 are benzyl and Ar is optionally substituted phenyl.

20. A compound according to claim 18 wherein Z^3 and Z^5 are toluyl and Ar is optionally substituted phenyl.

21. A compound according to claim 19 or 20 in which Ar is phenyl.

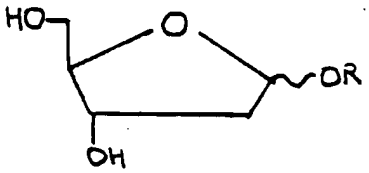
22. A method of treatment of virus infections which comprises administration to a recipient in need of treatment an effective amount of a compound according to claim 1.

23. A process for the preparation of a compound of the formula (XIII)



(XIII)

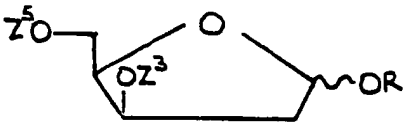
where R is a hydrocarbyl group and Z^3 and Z^5, which may be the same or different, are acyl groups, which comprises the reaction of a compound of the formula (XIV)



(XIV)

where R is as defined above, with a compound or compounds of formula Z^nOH where Z^n is Z^3 and/or Z^5.

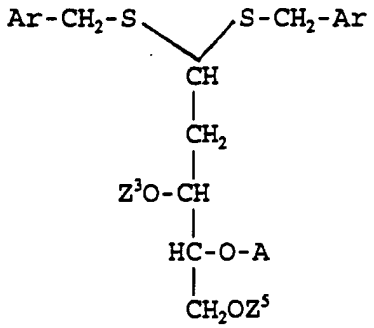
24. A compound of the formula (XIII)



(XIII)

where R is a hydrocarbyl group and Z^3 and Z^5, which may be the same or different, are acyl groups.

25. A compound of the formula (VII)

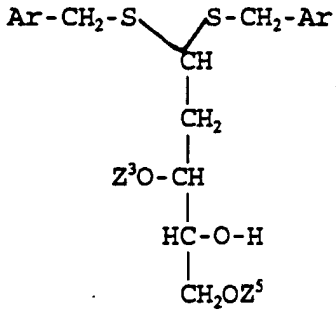


(VII)

where Z^3 and Z^5 are acyl groups, A is a leaving group and Ar is an optionally substituted aryl group.

26. A compound according to claim 25 wherein A is a methanesulphonyl group.

27. A compound of formula (VIII)



(VIII)

where Z^3 and Z^5 are acyl groups and Ar is an optionally substituted aryl group.

28. A compound according to any one of claims 25 to 27 wherein Ar is a phenyl or toluyl group optionally substituted by one or more halogen atoms, C_{1-4} alkyl eg. methyl, C_{1-4} haloalkyl, C_{1-4} alkoxy, nitro or amino groups.

29. A compound according to any one of claims 24 to 28, or a process according to claim 23 wherein the groups Z^3 and Z^5 are p-nitrobenzoyl.

INTERNATIONAL SEARCH REPORT

Inter. nal Application No
PCT/GB 93/01858

A. CLASSIFICATION OF SUBJECT MATTER
IPC 5 C07H19/06 A61K31/70

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 5 C07H A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO,A,90 01036 (MEDIVIR AB) 8 February 1990 see abstract ---	1-29
Y	EP,A,0 409 575 (THE UNIVERSITY OF BIRMINGHAM) 23 January 1991 cited in the application see abstract ---	1-29
Y	WO,A,91 04033 (SOUTHERN RESEARCH INSTITUTE) 4 April 1991 see page 3, line 3 - page 9, line 17 ---	1-29
Y	EP,A,0 421 777 (THE UNIVERSITY OF BIRMINGHAM) 10 April 1991 cited in the application see abstract ---	1-29
	-/--	

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
24 November 1993	0 3. 12. 93

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax (+31-70) 340-3016	Scott, J

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Inter nal Application No
PCT/GB 93/01858

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO,A,91 16333 (SOUTHERN RESEARCH INSTITUTE) 31 October 1991 see abstract	1-29
Y	WO,A,92 06993 (PASTEUR MERIEUX SERUMS ET VACCINS) 30 April 1992 see abstract	1-29
Y	WO,A,92 08727 (CONSIGLIO NAZIONALE DELLE RICERCHE) 29 May 1992 see page 1, line 1 - page 5, line 16	1-29
Y	WO,A,92 06102 (MEDIVIR AB) 16 April 1992 see page 1, line 1 - page 8, line 19	1-29

Form PCT/ISA/210 (continuation of second sheet) (July 1992)

International application No.
PCT/GB93/01858

INTERNATIONAL SEARCH REPORT

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Remark: Although claim 22 is directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searches without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

Int. mal Application No
PCT/GB 93/01858

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
WO-A-9001036	08-02-90	AU-A-	3978689	19-02-90
		EP-A-	0352248	24-01-90
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		WO-A-	9101326	07-02-91
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		EP-A-	0491793	01-07-92
		JP-T-	5500666	12-02-93
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		EP-A-	0506935	07-10-92
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WO-A-9206102	16-04-92	AU-A-	8641091	28-04-92
		CA-A-	2093020	03-04-92
		EP-A-	0554274	11-08-93